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PHILIPPINE CAMPHOR

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ONE PLATE

For centuries Formosa has been the world's greatest source of laurel camphor although considerable quantities are also exported from China and Japan. Owing to the increased demand for camphor and the new uses which have been found for it the cultivation has been extended to many parts of the world.

Some years ago camphor seeds obtained from Japan were planted in the Philippines. Although the camphor trees were not especially cultivated a number of them grew fairly well. Recently we have investigated the camphor content of many of these trees which are still young, and our experiments showed that in these trees the camphor is localized mostly in the leaves and not in the wood. While some of the trees yielded only camphor oil and others gave poor yields of camphor crystals, there were some trees which gave rather high yields of crystalline camphor. By careful selection and cultivation of the trees which gave high yields of crystals it might be possible to develop a natural-camphor industry which could compete successfully with the synthetic camphor now on the market. The results of our survey will supply certain basic data on the problem of natural-camphor production as it applies to the Philippines.

In considering the Philippine production of commercial camphor, it would seem advisable to investigate also the cultivation of Philippine (Baguio) pine trees and the turpentine obtained from them since synthetic camphor is now made from pine tree

turpentine. Aside from its use as a raw material for making synthetic camphor there is a considerable demand for turpentine as it is employed in the manufacture of high-grade paints and varnishes.

Although there are several varieties of camphor trees, the principal commercial species are the Borneo camphor, *Dryobalanops aromatica* Gaertner f., and the laurel camphor, *Cinnamomum camphora* Nees and Ebermaier. The Borneo camphor is obtained from trees grown in Sumatra, Borneo, and Java. Owing to the gradual depletion of the native forests and to the fact that reforestation has not been practiced to any extent, only a comparatively small quantity of Borneo camphor is now produced.[1] Laurel camphor is used principally in the manufacture of celluloid, celluloid products, and pharmaceutical preparations.

In the camphor-producing countries, such as Formosa and Japan, the laurel camphor is localized mostly in the wood of old forest trees while the leaves contain principally camphor oil.[2] Some of these forest trees are estimated to be over a hundred years old. The crude method of preparing camphor in these countries consists in cutting down the trees and chopping the wood into chips. The chips are then packed in perforated jars and heated over a crude steam bath. The steam enters the jars, saturates the chips, and causes the crude camphor to sublime and condense in earthenware pots placed over the jars. The crude camphor, thus prepared, contains a considerable amount of camphor oil which exudes from it. The oil is drained from the crystalline camphor which is refined by mixing it with quicklime and charcoal and subliming. The camphor oil contains a considerable amount of camphor which may be removed by distilling off a portion of the oil and freezing the camphor crystals out of the residue. The Japanese method of cutting down the trees and distilling the camphor from the wood is rather expensive as it requires a vast amount of reforestation to keep up the production.[3]

[1] Editorial, Phar. Journ. and Phar. 112 (1924) 234; Coblentz, V., Journ. Soc. Chem. Ind. 26 (1907) 382; Thorpe, E., Dictionary of Applied Chemistry 1 (1912) 615.

[2] Yearbook U. S. Dept. Agr. (1910) 455.

[3] Editorial, Phar. Journ. and Phar. 112 (1924) 235.

There is a considerable demand for camphor oil as it usually contains safrol which is used in making cheap perfumery, artificial oil of sassafras, heliotropin, and other products.[4] The ordinary camphor oil of commerce is a rather complex mixture of substances. When distilled, it may be separated into fractions of light and heavy oils. The light oil consists principally of terpenes and resembles oil of turpentine. The high-boiling, heavy oil contains sesquiterpenes and is usually rich in safrol. Camphor oils from different countries may vary greatly in composition. Oils from Mauritius, for instance, contain a considerable quantity of cineol but no safrol.[5]

After the Japanese took possession of Formosa the camphor industry became a government monopoly which controlled the production and price. As new uses were found for camphor there was naturally a much greater demand and the exportation from Japan and Formosa did not appear to be keeping up with the world's demand for this important commodity. Importers in other countries experienced considerable difficulty in obtaining sufficient supplies of camphor when they were required and there was frequently an uncertainty as to the price. As a result it was difficult for dealers to make close quotations for future deliveries. This situation naturally gave a great impetus to the commercial production of synthetic camphor, and, after many years of experimenting, this industrial chemical problem was finally worked out successfully. The raw material used for making synthetic camphor is pine tree turpentine,[6] which contains usually about 70 per cent of pinene. Numerous processes claiming to convert the pinene into camphor have been patented.[7]

Synthetic camphor is identical with natural camphor in all respects except that it is the inactive, racemic form of camphor and does not affect polarized light, whereas the natural camphor is optically active (dextro). The commercial value of synthetic camphor is shown by the fact that, since the year 1920, Germany has not imported any camphor and, moreover, has produced a

[4] Parry, E. J., The Chemistry of Essential Oils and Artificial Perfumes 1 (1918) 151–157; Editorial, Chem. Met. Eng. 22 (1920) 1030.

[5] Yearbook, U. S. Dept. Agr. (1910) 455.

[6] Editorial, Phar. Journ. and Phar. 112 (1924) 235.

[7] Pond, F. J., Journ. Soc. Chem. Ind. 26 (1907) 383; Editorial, Chemical Age 30 (1922) 211; Editorial, Chemist and Druggist, 105 (1926) 175.

surplus for export to the United States which consumes more camphor than any other country. In 1926 synthetic camphor was included in the new edition of the German Pharmacopœia thus making this product an official pharmaceutical preparation.[8] Synthetic camphor is now made not only in Germany but also in the United States and other countries.

The commercial success of synthetic camphor probably depends largely on the fact that the yield of turpentine pinene from pine trees is very much greater than the yield of camphor from camphor trees. Although it is rather costly to convert the pinene into camphor by synthetic chemical processes the expense is more than compensated by the comparatively large yields of camphor obtained. In the production of commercial camphor, it would seem that if the camphor trees are to compete successfully with the pine trees it will be necessary to increase and improve considerably the cultivation of camphor trees and, moreover, endeavor to develop strains that give much higher yields of camphor than are now usually obtained. A factor of advantage to the natural-camphor industry, however, is the valuable camphor oil which is obtained as a by-product. As yet no commercial synthetic substitute has been found for this oil.[9]

Pine trees (*Pinus insularis* Endlicher) grow well in the Philippines in the highlands of central and northern Luzon [10] at altitudes varying from about 900 to 1,500 meters. Turpentine obtained from Philippine pine trees has been investigated by Richmond [11] and also by Brooks.[12] The trees selected for their investigations were growing near Baguio, a summer resort situated at an elevation of about 1,500 meters in the mountain province of the Philippines. Their results showed that some of the trees gave fair yields of turpentine while others gave rather poor yields. According to Brooks, turpentine from the Baguio pine trees consists principally of pinene and compares favorably with American turpentine. When time permits we expect to make a survey of the turpentine resources of the Philippines

[8] Editorial, Chemist and Druggist 105 (1926) 604.
[9] Editorial, Phar. Journ. and Phar. 112 (1924) 235.
[10] West, A. P., and W. H. Brown, Philip. Bur. Forestry Bull. 20 (1920) 32.
[11] Philip. Journ. Sci. § A 4 (1909) 231.
[12] Philip. Journ. Sci. § A 5 (1910) 229.

since turpentine is not only useful for making synthetic camphor but is also employed in the manufacture of high-grade paints and varnishes.

In 1910 the Philippine Bureau of Forestry began the cultivation of laurel camphor in the Philippines. Seeds were obtained from Japan and planted at the forestry nursery in Baguio. Since then a number of trees have been planted at different times in various parts of Baguio and at the Trinidad Farm School which is nearby. Compared to old forest trees in Formosa these Philippine trees are really very young. Considering that the trees were not really cultivated, many of them have grown fairly well both from seeds and from cuttings. The forester, S. Laraya, in charge of the forestry nursery, estimates that there are now over two thousand camphor trees in Baguio and the vicinity. These trees are located approximately as follows: About 1,700 trees in Forbes Park, about 60 along the Pack Road and South Drive, and about 400 at the Trinidad Farm School. These trees range in height from about 2 meters for the younger trees to about 13 meters for the older ones. Leaves on many of the older trees were found to be infected with a kind of fungus growth. With proper cultivation this plant disease could probably be prevented.

EXPERIMENTAL PROCEDURE

During the spring of 1929 we decided to make a general survey of the camphor situation in Baguio. Through the courtesy of the Commanding Officer of Camp John Hay we obtained permission to use the plumber's shop at this military post as a field laboratory. The army officers very kindly provided us with electric lights and laboratory tables. All necessary chemical supplies and equipment were transported from the Bureau of Science in Manila to this field laboratory in Baguio.

The trees selected for our investigation were numbered, and a metal label containing the number was attached firmly to each tree for future identification. The data concerning these trees are recorded in Table 1, which gives the number of each tree, approximate location, and conditions of growth, such as height and position (shady or sunny) on hillside or elsewhere.

As shown by the data (Table 1), we selected for our investigation camphor trees of various sizes and growing under different conditions.

TABLE 1.—*Location and description of Philippine trees selected for distilling camphor.*

Tree No.	Location.	Height.	Position.	Light.
	Forest nursery:	*Meters.*		
2	South of office	6.5	Ravine	Half shady.
2-A	do	5.5	do	Do.
2-B	do	7.0	do	Shady.
2-C	do	3.5	do	Sunny.
3	do	4.5	do	Do.
3-A	do	4.0	do	Half shady.
3-B	do	3.5	do	Do.
3-C	do	3.5	do	Do.
4	do	1.5	do	Do.
4-A	do	2.0	do	Sunny.
4-B	do	1.5	do	Shady.
4-C	do	2.5	do	Half shady.
4-D	do	3.5	do	Do.
11	Below ranger's house	13.5	do	Do.
11-A	do	12.0	do	Do.
11-B	do	3.5	do	Do.
14	West of office	6.5	Hillside	Sunny.
14-A	do	6.0	do	Do.
14-B	do	7.0	do	Do.
14-C	do	6.5	do	Do.
14-D	do	6.0	do	Do.
14-E	do	6.0	Ravine	Sunny.
14-F	do	3.5	do	Do.
15-B	do	4.0	do	Half shady.
15-C	do	5.0	do	Do.
15-D	do	3.0	do	Sunny.
15-E	do	3.0	Hillside	Do.
15-F	do	5.0	do	Half shady.
16-A	Southwest of office	6.0	Ravine	Sunny.
16-B	do	7.0	do	Do.
5	Italian garden	10.5	Level land	Half shady.
5-A	do	9.5	Ravine	Do.
5-B	do	7.0	do	Shady.
5-C	do	9.0	do	Do.
5-D	do	9.5	do	Half shady.
5-E	do	10.0	do	Do.
	South drive:			
7	Near Mission House	4.5	Hillside	Do.
7-B	do	5.5	do	Half shady.
7-C	do	3.5	do	Do.
7-D	do	3.1	do	Half shady.
8	do	9.0	do	Do.
8-A	Near Teacher's Camp	9.0	do	Shady.
8-B	do	9.0	do	Do.
8-C	do	9.0	do	Do.
8-D	do	9.0	do	Do.
8-E	do	10.0	do	Do.
8-F	do	10.0	do	Half shady.
8-G	do	4.5	do	Do.

TABLE 1.—*Location and description of Philippine trees selected for distilling camphor*—Continued.

Tree No.	Location.	Hieght.	Position.	Light.
	Teacher's Camp:	*Meters.*		
6	Near Barrows Hall	11.0	Hill	Sunny.
6-A	do	6.5	do	Do.
6-B	do	5.5	do	Half shady.
9	Below suspension bridge	10.0	Ravine	Do.
9-A	do	6.5	do	Do.
9-B	do	8.2	do	Do.
9-C	do	1.5	do	Shady.
9-D	do	1.4	do	Do.
9-E	do	5.0	do	Half shady.
9-F	do	6.2	do	Do.
10	do	5.0	do	Do.
10-A	do	9.0	do	Do.
10-B	do	6.2	do	Do.
10-C	do	4.8	do	Do.
10-D	do	4.0	do	Do.
10-E	do	1.2	do	Do.
10-F	do	1.8	do	Do.
12	Session Road near Military Circle	4.0	Hillside	Do.
12-A	do	1.8	do	Do.
12-B	do	3.5	do	Do.
12-D	do	3.1	do	Do.
12-E	do	2.2	do	Do.
12-G	do	2.1	do	Do.
12-H	do	2.5	do	Do.
12-I	do	1.7	do	Shady.
12-J	do	2.0	do	Do.
13	do	2.2	do	Do.
13-A	do	1.6	do	Sunny.
13-B	do	2.0	do	Do.
13-C	do	2.6	do	Half shady.
17-A	Bureau of Agriculture Experiment Station	2.5	Ravine	Sunny.
17-B	do	3.0	do	Do.
17-C	do	3.0	do	Do.
17-D	do	2.0	do	Do.
	Pack Road:			
18	Near Baguio station	4.5	Hillside	Half shady.
18-A	do	3.0	do	Sunny.
18-B	do	3.0	do	Shady.
18-C	Opposite Pines Hotel	2.5	do	Do.
18-D	do	3.0	do	Do.
18-E	do	3.0	do	Half shady.

Before beginning our routine work on determining the camphor content of Philippine camphor trees we carried out a number of preliminary experiments relating to the moisture, oil, and crystal camphor in the trees.

In Baguio, during the late springtime, afternoon and evening showers occur frequently. The moisture content of moist, damp camphor leaves gathered early in the morning is greater than that of leaves gathered later in the day when the sun is overhead and the temperature is much higher than in the early morning. If the camphor content of the leaves is calculated on the weight of the green leaves then, due to the moisture present, the percentage of camphor in leaves from the same tree will vary according to the time the leaves are gathered, whether in the early morning or at noon. In order to have a definite standard for comparison it would seem that the crystal camphor content of the leaves should be calculated on a moisture and oil free basis. It should make practically no difference then at what time of the day the leaves are gathered as leaves from the same tree should always give approximtely the same camphor content.

When the usual method for determining moisture is applied to camphor leaves we find that the leaves, when heated in an oven, lose not only moisture but also the volatile camphor and camphor oil. When the camphor leaves are ground to a pulp and heated long enough they lose all their moisture and also all the camphor and camphor oil. This is proved by the fact that the odor of camphor or camphor oil cannot be detected in the dried residue and also when the residue is distilled with water no camphor crystals or oil passes over into the distillate.

In calculating the results of our experiments we have based our calculations both on the weight of moist green material and also on a moisture and oil free basis. Our experimental procedure was as follows: A quantity of camphor leaves from a particular tree was ground in a meat grinder. The pulp was mixed thoroughly and a sample (100 grams) treated with water and distilled about two hours. According to our experiments this is a sufficient length of time to remove all the camphor from 100 grams of leaves. The camphor crystals obtained from the aqueous distillate and from the cooled portion of the Liebig condenser were pressed between layers of filter paper, dried in a desiccator, and then weighed. A sample of ground leaves (2 grams) was heated in an oven at a temperature of about 95° C. to constant weight. This required usually about seven hours. The loss in weight represented the moisture, crystal camphor, and camphor oil. The results were then calculated on the basis

of 100 grams of leaves. The following notes give a summary of one of our experiments.

100 grams of leaves distilled gave 1.67 grams of camphor.
100 grams of leaves dried to constant weight gave a loss in weight of 51.45 grams.

100.00 grams of camphor leaves.
51.45 moisture, crude crystal camphor, and volatile oil.

48.55 nonvolatile solids in 100 grams of leaves.

51.45
1.67 crude crystal camphor.

49.78 moisture and oil in 100 grams of leaves.

100.00
49.78

50.22 crude crystal camphor and nonvolatile solids in 100 grams of moist, green leaves.

$\frac{1.67 \times 100}{50.20} = 3.32$ percentage of crude crystal camphor calculated on a moisture and oil free basis.

Analysis of camphor leaves calculated on the weight of moist green leaves would then be as follows:

	Per cent.
Crystal camphor	1.67
Moisture and oil	49.78
Nonvolatile solids	48.55
Total	100.00

The crude camphor crystals usually contain a very small amount of oil which slightly increases the weight. According to our experiments 100 grams of the crude crystals contain usually about 1 gram of oil. This slight increase in the weight of the crystals due to the oil present is about equivalent to the small amount of camphor which is probably volatilized and lost in the distillation and drying of the crystals. Our figures, showing the percentage of crude camphor calculated on a moisture and oil free basis, seem to be very nearly correct.

The camphor in young Philippine trees seems to be located almost entirely in the leaves, for when the wood and twigs of Philippine camphor trees are distilled only traces or very small amounts of camphor crystals are obtained. This fact is brought out very clearly by the data given in Table 2. Possibly, if these

Philippine trees grew to an old age the camphor content of the wood might compare favorably with that of old forest trees in Formosa.

TABLE 2.—*Crystal camphor in leaves, stems, and small branches of Philippine trees.*

Tree No.	Height of tree.	Part of tree.	Analysis.			
			Moisture and oil.	Nonvolatile solids.	Crystal camphor in moist material.	Crystal camphor calculated on moisture and oil free basis.
	Meters.		*Per cent.*	*Per cent.*	*Per cent.*	*Per cent.*
3–B	3.5	Leaves	66.04	32.56	1.40	4.12
3–B	3.5	Stems	67.16	32.61	0.23	0.70
3–B	3.5	Small branches	54.26	45.74	trace	trace
4	1.5	Leaves	60.09	38.91	1.00	2.50
4	1.5	Stems	65.66	34.14	0.20	0.58
4	1.5	Small branches	52.03	47.97	trace	trace
4–B	1.5	Leaves	54.46	43.85	1.69	3.71
4–B	1.5	Stems	60.63	39.27	0.10	0.25
4–B	1.5	Small branches	51.90	48.10	trace	trace
5	10.5	Leaves	56.96	40.95	2.09	4.86
5	10.5	Stems	62.30	36.91	0.79	2.10
5	10.5	Small branches	51.57	48.26	0.17	0.35
6	11.0	Leaves	55.72	43.16	1.12	2.53
6	11.0	Stems	57.73	42.11	0.16	0.38
6	11.0	Small branches	51.28	48.67	0.05	0.10
6–A	6.5	Leaves	60.96	37.85	1.19	3.05
6–A	6.5	Stems	62.64	37.14	0.22	0.59
6–A	6.5	Small branches	51.30	48.69	0.01	0.02

The data (Table 2) show also the difference between calculating the yield of crystal camphor on the weight of moist, green leaves and on a moisture and oil free basis. When calculated on the weight of moist, green material, the leaves of tree 3–B gave a smaller yield of crystal camphor (1.40 per cent) than the leaves of tree 4–B, which gave 1.69 per cent. When calculated on a moisture and oil free basis the leaves of tree 3–B gave a higher yield of camphor (4.12 per cent) than the leaves of tree 4–B (3.71 per cent). Since many of the analyses recorded in Table 3 also gave this kind of results, it would seem that the correct standard for comparing the camphor content of different trees would be to calculate the results on a moisture and oil free basis.

Since the camphor in Philippine trees seems to be located mostly in the leaves, our investigation consisted principally in distilling the leaves of a large number of trees and determining

the yield of crystal camphor in these leaves. The results are recorded in Table 3.

As shown by the data (Table 3) the leaves of Philippine camphor trees gave about 2 to 7 per cent of crystal camphor calculated on a moisture and oil free basis. This may seem to be a wide variation, but according to Rusby [13] the yield of camphor from trees in any particular locality may vary so much that it is rather difficult to estimate an average yield. This may be due to the fact that camphor is formed in the plant by certain physiologic processes and the natural synthesis may, perhaps, be very sensitive to the inherent characteristics of certain strains and also to slight environmental changes.

Data on the Philippine trees which gave the highest yields of crystal camphor are given in Table 4. By careful cultivation of these particular trees it might be possible to develop a natural-camphor industry which could compete successfully with the

TABLE 3.—*Crystal camphor in leaves of Philippine trees.*

Tree No.	Height of tree.	Analysis.			
		Moisture and oil.	Nonvolatile solids.	Crystal camphor in green leaves.	Crystal camphor calculated on moisture and oil free basis.
	Meters.	*Per cent.*	*Per cent.*	*Per cent.*	*Per cent.*
2	6.5	49.81	48.09	2.10	4.18
2-A	5.5	66.14	31.97	1.89	5.58
2-B	7.0	50.26	48.04	1.70	3.42
2-C	3.5	62.66	36.22	1.12	3.00
3	4.5	50.67	47.13	2.20	4.46
3-A	4.0	64.31	34.18	1.51	4.23
3-C	3.5	66.72	31.67	1.61	4.84
4-A	2.0	60.96	37.63	1.41	3.61
4-C	2.5	63.17	35.95	0.88	2.39
4-D	3.5	64.81	34.00	1.19	3.38
5-A	9.5	60.06	38.13	1.81	4.53
5-B	7.0	66.74	31.75	1.51	4.54
6-B	5.5	55.07	43.86	1.07	2.38
7	4.5	55.68	42.67	1.65	3.72
7-B	5.5	55.52	43.18	1.30	2.92
7-C	3.5	64.42	34.77	0.81	2.28
7-D	3.1	54.74	44.39	0.87	1.92
8-B	9.0	54.53	44.22	1.25	2.75
8-C	9.0	56.65	41.93	1.42	3.28
8-D	9.0	59.83	38.79	1.38	3.44
8-E	10.0	58.11	40.51	1.38	3.29
9	10.0	58.51	39.06	2.43	5.86
9-A	6.5	60.50	37.60	1.90	4.81

[13] Journ. Soc. Chem. Ind. 26 (1907) 381.

TABLE 3.—*Crystal camphor in leaves of Philippine trees*—Continued.

Tree No.	Height of tree.	Analysis.			
		Moisture and oil.	Nonvolatile solids.	Crystal camphor in green leaves.	Crystal camphor calculated on moisture and oil free basis.
	Meters.	*Per cent.*	*Per cent.*	*Per cent.*	*Per cent.*
9–B	8.2	57.59	40.86	1.55	3.65
9–C	1.5	62.19	35.72	2.09	5.53
9–D	1.4	62.59	35.51	1.90	5.08
9–F	6.2	60.70	37.45	1.85	4.71
10	5.0	60.57	37.97	1.46	3.70
10–A	9.0	54.91	43.50	1.59	3.53
10–B	6.2	57.27	41.39	1.34	3.14
10–C	4.8	55.29	41.93	2.78	6.22
10–D	4.0	62.95	35.28	1.77	4.78
10–E	1.2	60.47	37.67	1.86	4.71
10–F	1.8	59.07	39.20	1.73	4.23
11–A	12.0	62.70	35.72	1.58	4.24
11–B	3.5	67.38	31.28	1.34	4.11
12	4.0	63.42	34.88	1.70	4.65
12–A	1.8	60.40	37.35	2.25	5.68
12–B	3.5	64.96	32.98	2.06	5.88
12–D	3.1	60.69	36.36	2.95	7.50
12–E	2.2	65.15	32.45	2.40	6.89
12–H	2.5	58.69	38.43	2.88	6.97
12–J	2.0	60.20	37.47	2.33	5.85
13	2.2	57.05	40.37	2.58	6.01
13–A	1.6	64.19	33.84	1.97	5.50
13–B	2.0	63.01	34.23	2.76	7.46
13–C	2.6	61.74	36.08	2.18	5.70
14	6.5	60.18	37.38	2.44	6.13
14–A	6.0	57.12	40.53	2.35	5.48
14–B	7.0	61.95	36.25	1.80	4.73
14–C	6.5	72.52	25.75	1.73	6.30
14–D	6.0	66.66	31.59	1.75	5.25
14–E	6.0	64.99	33.62	1.39	3.97
14–F	3.5	65.15	33.82	1.03	2.95
15–B	4.0	60.03	38.83	1.14	2.85
15–D	3.0	64.75	33.78	1.47	4.17
15–E	3.0	62.85	35.14	2.01	5.41
16–A	6.0	63.01	35.17	1.82	4.92
17–A	2.5	63.37	35.47	1.16	3.17
17–B	3.0	61.37	36.31	2.32	6.01
17–C	3.0	64.41	33.84	1.75	4.92
17–D	2.0	62.58	35.35	2.07	5.53
18	4.5	56.74	41.26	2.00	4.62
18–A	3.0	57.11	41.33	1.56	3.64
18–B	3.0	55.56	41.65	2.79	6.28
18–C	2.5	58.15	39.69	2.16	5.16
18–D	3.0	55.21	41.65	3.14	7.01
18–E	3.0	59.53	39.11	1.36	3.36

synthetic camphor now on the market. Possibly, if these trees were cultivated in other parts of the Philippines where the soil conditions are different, as in Bukidnon, Mindanao, the yield of crystal camphor might be higher than in Baguio.

TABLE 4.—*Philippine trees which gave the highest yields of crystal camphor.*

Tree No	Location.	Height.	Position.	Light.	Crystal camphor calculated on moisture and oil free basis.
	Teacher's camp:	*Meters.*			*Per cent.*
9	Below suspension bridge	10.0	Ravine	Half shady	5.86
10–C	do	4.8	do	do	6.22
12–A	Sessions Road near Military Circle	1.8	Hillside	do	5.68
12–B	do	3.5	do	do	5.88
12–D	do	3.1	do	do	7.50
12–E	do	2.2	do	do	6.89
12–H	do	2.5	do	do	6.97
12–J	do	2.0	do	do	5.85
13	do	2.2	do	do	6.01
13–B	do	2.0	do	Sunny	7.46
13–C	do	2.6	do	Half shady	5.70
14	Forest nursery west of office	6.5	do	Sunny	6.13
14–C	do	6.5	do	do	6.30
17–B	Bureau of Agriculture Experiment Station.	3.0	Ravine	do	6.01
	Pack Road:				
18–B	Near Baguio Station	3.0	Hillside	Shady	6.28
18–D	Opposite Pines Hotel	3.0	do	do	7.01

Crystal camphor obtained by distilling Philippine leaves is always accompanied by a small quantity of light yellow camphor oil which amounts to about 1 per cent of the weight of camphor crystals. In addition to this small amount of oil which adheres to the crystals there is a very small quantity of a slightly yellow volatile oil that passes through the condenser into the aqueous distillate. This amounts to about 0.22 per cent calculated on the weight of green leaves. As the yield of this volatile oil was so small we did not prepare a sufficient amount for investigation. Our results seem to indicate that crystal camphor is the principal product obtained by distilling Philippine camphor leaves and only a very small quantity of camphor oil is obtained as a by-product.

The crude Philippine camphor was washed with warm water to remove the oil which adhered to the crystals. The crystals were then dried in a desiccator. The melting point of the crystals was found to be 174.5 to 175.5° C.

Watts [14] gives 175° C. as the melting point of pure camphor.

According to Thurston,[15] a solution prepared by dissolving 10 grams of camphor in 100 cubic centimeters of alcohol (95 per cent) gives at 25° C. in a 200-millimeter tube a specific rotation $(A \frac{25° C.}{D})$ of +41 to +42°. Under the same conditions but at a slightly different temperature the Philippine camphor gave a specific rotation $(A \frac{28.5° C.}{D})$ of +41.62°.

The camphor oil, which was removed from the crude crystals by warm water, was extracted from the aqueous solution with ether. The oil was found to have a specific rotation $(A \frac{30° C.}{D})$ of +26.01°.

The leaves of some Philippine camphor trees, when distilled, yielded no crystal camphor but only camphor oil. Analyses of leaves from trees which gave only camphor oil are recorded in Table 5. These results were calculated in a manner similar to that employed in calculating the yield of crystal camphor on a moisture and oil free basis. In general, the yield of camphor oil is less than the yield of crystal camphor.

TABLE 5.—*Camphor oil in leaves of Philippine trees.*

Tree No.	Height of tree.	Analysis.			
		Moisture.	Nonvolatile solids.	Camphor oil in green leaves.	Camphor oil calculated on moisture free basis.
	Meters.	*Per cent.*	*Per cent.*	*Per cent.*	*Per cent.*
7–A	4.5	56.74	42.04	1.22	2.82
8–F	10.0	52.49	45.82	1.69	3.56
9–E	5.0	57.91	40.70	1.39	3.30
11	13.5	56.12	42.44	1.44	3.28
12–G	2.1	57.91	38.88	3.21	7.63
12–I	1.7	68.26	31.08	0.66	2.08
15–C	5.0	66.40	33.34	0.26	0.77
15–F	5.0	54.81	43.68	1.51	3.34
16–B	7.0	55.61	42.54	1.85	4.17

[14] Dictionary of Chemistry 1 (1927) 668.

[15] Pharmaceutical and Food Analysis (1922) 5.

This slightly yellow camphor oil, which was obtained from leaves that gave no crystals, was found to have the following constants:

Specific gravity $(d \frac{30^\circ C.}{30^\circ C.})$, 0.8858

Refractive index $(N \frac{29.5^\circ C.}{D})$, 1.4652

Specific rotation $(A \frac{30^\circ C.}{D})$, —19.2 °

Since this oil had a negative rotation (—19.2 °) it is evidently quite different from the ordinary commercial camphor oil which has a positive rotation. Some years ago the leaves of some camphor trees in Mauritius were distilled and the distillate was found to contain no crystal camphor but only camphor oil which gave a specific rotation [16] of —20.4°. It is said that in Formosa there are also varieties of trees which yield no crystal camphor but only camphor oil. Possibly this unusual Philippine oil which had a negative rotation is similar to the oil obtained from leaves in Mauritius. We expect to investigate the composition of this Philippine oil when a sufficient supply is available.

SUMMARY

We have investigated a large number of Philippine camphor trees growing at Baguio in the mountain province of the Philippines. These trees were of various sizes and were growing under different conditions. The results of our experiments have shown that camphor in young Philippine trees is located almost entirely in the leaves, for when the wood and twigs are distilled only traces or very small amounts of camphor crystals are obtained.

The leaves of Philippine camphor trees gave about 2 to 7 per cent of crystal camphor calculated on a moisture and oil free basis. The yield of camphor was also calculated on the weight of moist green leaves, but, as shown by our figures, the results are apt to be misleading due to the variable amount of moisture contained in the leaves.

By careful cultivation of the particular trees which gave the highest yields of crystal camphor it might be possible to develop a natural camphor industry in the Philippines. Possibly, if these trees were cultivated in other parts of the Philippines where the soil conditions are different the yield of crystal camphor might be higher than in Baguio.

[16] Parry, E. J., Essential Oils and Artificial Perfumes (1918) 157.

High-grade camphor may be obtained from the leaves of Philippine trees, but those trees which yielded crystal camphor gave only a small quantity of camphor oil as a by-product.

The leaves of some Philippine trees, when distilled, yielded no crystal camphor but only camphor oil which had a negative rotation. Similar results were obtained from leaves of camphor trees in Mauritius. These oils which have a negative rotation are different from the ordinary commercial camphor oil which has a positive rotation.

In considering the Philippine production of commercial camphor it would seem advisable to investigate the cultivation of Philippine pine trees and the turpentine obtained from them. Aside from its use as a raw material for making synthetic camphor there is a considerable demand for turpentine as it is also employed in the manufacture of high-grade paints and varnishes.

ILLUSTRATION

PLATE 1. Philippine camphor trees in Baguio.

PLATE 1. PHILIPPINE CAMPHOR TREES IN BAGUIO.

BENDING AND COMPRESSIVE STRENGTHS OF THE COMMON PHILIPPINE BAMBOO [1]

By José C. Espinosa

Of the Bureau of Science, Manila

THREE PLATES

INTRODUCTION

One of the most important building materials in the Philippines is the common bamboo, *Bambusa spinosa* Roxburgh. Much has been written about its varied usefulness in tropical countries; and to quote Brown and Fischer:[2]

> The domestic uses of bamboo are innumerable and include bridges, fences, rafts, fish-traps, vessels for carrying and storing water, cooking, splints for baskets, hats and mats, vehicle shafts, chairs, cupboards, tables, beds, benches, flower pots, etc. In fact on account of the ease with which it is worked, bamboo is used for almost every purpose for which wood is employed in temperate countries.

Houses of light material are generally of bamboo and nipa, and for temporary military bridges in tropical countries, bamboo is unsurpassed in availability of material, lightness, ease of construction, and strength.

The present consideration is limited to the bending and compressive strengths of the common Philippine bamboo, *Bambusa spinosa* Roxburgh, the most useful of the thirty species known to be found in the Islands.

SAMPLES TESTED

The samples used in these tests were obtained in the local market where gradings due to size have placed them in the first-class lots. They were cut after the rainy season, or in the months of November and December, and are representative of the common Philippine bamboo used in building construction. They were subsequently seasoned and tested in the latter part

[1] This work has been done in coöperation with the Bureau of Forestry by the aid of their representative, Ranger Martin Lagrimas, who made the tests under the supervision of the author.

[2] Bull. Philip. Bur. of Forestry 15 (1918) 10.

of March and the first half of April following. They were, therefore, dry and free from sap. Bamboo cut during the rainy season generally contains sap and is easily attacked by insects, especially "boc-boc," a species of boring beetle.

The specimens were from 10 to 15 meters in length with a culm diameter of about 10 centimeters. From each specimen the following test pieces were obtained: Starting from the culm end, one piece including five nodes for bending tests of 5-foot span; one piece with three nodes, and when the joints are shorter five nodes, and 48 inches high, for compression parallel to the grain or column test; two or three pieces with three nodes for bending tests of 28-inch span; and three pieces with two internodes and 14 inches high for compression parallel to the grain. Smaller test pieces of split bamboo were cut from about the middle of each specimen. For bending tests, the pieces were about 9 by 15 millimeters in cross section and long enough for a span of 12 inches. The compression pieces were 1 inch high, the cross section being about the same as the bending pieces. The first test may be called, split-bamboo bending, and the second, split-bamboo compression.

TESTING MACHINE AND ACCESSORIES

All these tests were made on an Olsen testing machine of 30,000 pounds capacity. A deflectometer was used with which readings of deflection may be taken to 0.005 of an inch. Different speeds were used in bending and compression as may be seen in the following tests.

METHOD OF TEST

Static bending; 5-foot span.—The test piece in this case was supported on the outer nodes. Where the outer nodes do not come exactly on the points of support, the specimen was arranged so that these nodes are equidistant from the supports. Only test pieces of about the same size and approximately the same length of internodes were used in this test.

Load was gradually applied at the center node, the span being 60 inches. The head of the machine moved at the rate of 0.225 inch per minute. Readings of deflection were taken for every 100-pound increase in load (Table 1).

Column test.—The object of the test was to find the strength of bamboo posts as generally used in Filipino construction. One end of the specimen was U-shaped to take rounded bearing surface; this is the common practice among bamboo carpenters.

The other end was cut at right angles to the axis. Plate 2 shows the manner of loading; "W" is a piece of wood which exactly fits the U end. The speed of the machine in this case was 0.042 inch per minute.

Static bending; 28-inch span.—The object of this test was to determine the strength of bamboo when used as floor joists either in house or bridge construction. Deflection readings were taken for every 100-pound increase in load. The speed of the machine was 0.225 inch per minute.

Compression parallel to the grain.—The specimen in this case included only two internodes and was cut about 1 inch from each node at right angles to the axis. The values to be determined were those occurring in practice where short posts are used, as in low platforms and floors. Load was applied parallel to the grain until the specimen failed. The speed of the machine was 0.042 inch per minute.

Split-bamboo bending.—This test had for its object, the determination of the strength of split bamboo when used as flooring or wattling. The test pieces included only the internodes. Loading was applied at the center. Spans of 12 inches were used; forty-three pieces were bent with the exterior side up, and an equal number with the interior side up. Loading was gradual, and for every increase of 10 pounds a deflection reading was taken. The greatest fiber stress at the instant of failure was computed from the formula $f = \frac{M\,y}{I}$ where f is the fiber stress, M is the maximum moment, y is one-half the height, and I is the moment of inertia of the section along its intersection with the neutral plane.

Split-bamboo compression.—In order to have an idea of the crushing strength parallel to the grain of bamboo, compared with wood and other building materials this test was carried out. Load was applied on the ends of the specimen, 1 inch high, until failure occurred.

MANNER OF FAILURE AND POINTS OF INTEREST OBSERVED

Invariably in bending tests of both the large and small specimens failure was due to splitting along the sides and top, starting from the center and running outward in both directions. Upon releasing the load after a test, the specimen returned to normal so perfectly that it was difficult to recognize the marks left by the points of support and application of load. A thin slit may remain where splitting occurred.

Likewise in the compression tests splitting was the cause of failure. This, however, was generally accompanied by compression. As might be expected in the case of the larger specimens with the U-shaped top ends, splitting began from the top, but failure due to compression also appeared at the lower end where there was a general separation of the fibers as in a brush. In the case of the smaller specimens failures due to both splitting and compression, as well as general fiber separation, occurred at both ends.

Split bamboo in bending behaved somewhat differently. When the specimens were bent with the exterior side up, failure was due to tension on the lower side and to longitudinal shear. The maximum total deflection observed was 0.6 inch. Hardly any mark was left at the point where the load was applied. On the other hand, in bending with the exterior side down failure was due to longitudinal shear, as well as to actual crushing of the specimen where the load was applied, and, ultimately, tension on the exterior side. It may be of interest to note that the maximum load was carried for a total deflection of, in some cases, 3 inches without injury to the exterior side, indicating the great flexibility of the material under test.

The following are the values for greatest fiber stress at the instant of failure.

	Exterior side up.	Exterior side down.
Maximum	2,160	1,830
Average	1,430	1,130
Minimum	885	689

Bond [3] found that from twenty small beams tested, there was no difference in strength observable when specimens were loaded with the exterior side up and with the exterior side down.

Failure in the case of split-bamboo compression was due both to compression and general separation of the fibers at both ends and to splitting.

The following crushing strengths were found: Maximum, 749; average, 535; minimum, 352.

DISCUSSION

In this connection it is probably not out of place to recall the structure of bamboo so as to have a better understanding whereof bamboo owes its unusual strength for its size. Plate 3

[3] Professional Memoirs, Corps of Engineers, United States Army 5: 601.

shows an enlargement [4] of a cross section of a bamboo segment. It can be seen how close together the hard sclerenchyma cells are arranged toward the exterior side, and that they become more and more dispersed toward the inside. They are so close together toward the exterior side that the parenchyma cells, pronounced on the inside, are practically absent. The exterior itself is hard. Determinations of the hardness of the exterior by means of a scleroscope showed that bamboo approximates mild steel, giving an average reading of 30. Thus we have in bamboo a hollow cylinder the wall of which is composed of long straight fibrovascular bundles with an outside casing of a very hard material.

It can be seen that the strength tests in bending and compression in the case of bamboo have variations which cannot be termed within narrow limits. On the other hand these limits are not so wide but that the data lend themselves to generalizations sufficiently accurate for engineering purposes. It should be remembered that the specimens herein used varied in thickness, size, length of internodes, and straightness of growth. We were, therefore, confronted with innumerable fcators affecting the strengths.

In Tables 1 and 2 are given the results of tests in static bending for spans of 60 inches and 28 inches, respectively. The strength values are herein grouped according to thickness of wall and according to outside circumference for the same thickness of wall. It will be shown in the summary following that the strength values vary with the thickness of wall. From the last column of Tables 1 and 2, it would seem that in general for specimens of the same thickness of wall, the strength increases with an increase in outside circumference. This is true where a large number of tests have been made.

In Table 3 is given a summary of the results of tests in static bending with the specimen supported near the ends and loaded at the center. The figures for the specimens are grouped according to thickness. Average values of crushing loads for different thicknesses with the corresponding average outside circumferences are here tabulated. For both 60- and 28-inch

[4] Courtesy of L. J. Reyes, of the Bureau of Forestry, who furnished the original photograph and aided the author in the explanation of the bamboo structure.

spans it can be seen that the size of the specimens varied little and that an increase in thickness is accompanied by a material increase in strength as shown by the average values of crushing load. This is to be expected, and, while it is unfortunate that on account of several other factors like length of internodes, kind of nodes, and straightness of growth, no definite mathematical relationship can be derived from these data, it is safe to assume that a piece of common bamboo of about 30 centimeters in outside circumference can take a transversal live load of 0.25 to 1 ton, the span being 5 feet, before crushing occurs. Smaller pieces of about 24 centimeters outside circumference will take a live load of 0.25 to 0.75 ton if the span is about 2 feet.

A few specimens were broken with the supports and the load at points midway between the nodes. The strength is below 50 per cent of the above values. The practice, however, is to support a beam at the node where it is shown to be more resistant to crushing.

Tables 4 and 5 are the results of tests in compression parallel to the grain for column and for short posts, respectively. They are herein grouped according to thickness of wall and to outside circumference for the same thickness of wall.

Table 6 is a summary of the strength that may be expected when bamboo is loaded parallel to the longitudinal axis. In the column tests it can be seen that a thicker bamboo should give higher strength values. The relationship of thickness to strength is not in direct proportion because increase in bearing surface alone due to greater thickness is not the only factor affecting strength. This is clearly shown by the tests on smaller specimens, in that the average strength value for specimens 5 millimeters in thickness is slightly lower than that for specimens 4 millimeters in thickness.

For bamboo columns 4 feet high a minimum value of 4,630 kilograms and a maximum strength of 12,300 kilograms or from 4.5 to 12 tons have been registered. Ultimate failure occurred with a sudden drop in strength; the fibers, however, held together even at this point, except where the actual failure took place.

The smaller compression specimens, about 7 centimeters in diameter and 14 inches high, in general can support 4.5 tons, and may support a load of as much as 8.5 tons.

TABLE 1.—*Static bending.*

[Span, 60 inches; beam loaded at center with the points of support and application of load at or near the nodes; speed of machine, 0.225 inch per minute.]

Average thickness of wall.	Average outside circumference.	Crushing load.	Average crushing load for the same thickness of wall and the same outside circumference.	Average thickness of wall.	Average outside circumference.	Crushing load.	Average crushing load for the same thickness of wall and the same outside circumference.
mm.	*cm.*	*kg.*	*kg.*	*mm.*	*cm.*	*kg.*	*kg.*
5	28	442		7	30	447	
5	28	442		7	30	668	
5	28	611	498	7	30	706	
5	29	351		7	30	725	637
5	29	396	373	7	31	446	
5	30	573	573	7	31	513	
5	31	351	351	7	31	581	
6	28	294		7	31	713	
6	28	394		7	31	736	
6	28	441		7	31	779	
6	28	450		7	31	805	
6	28	467		7	31	900	684
6	28	543		7	32	509	
6	28	609		7	32	573	
6	28	670	484	7	32	611	
6	29	283		7	32	623	
6	29	351		7	32	827	629
6	29	373		7	33	633	
6	29	385		7	33	690	667
6	29	430		7	34	602	602
6	29	442		8	28	668	
6	29	475		8	29	375	
6	29	570		8	29	482	429
6	29	528		8	30	792	
6	29	573		8	31	573	
6	29	597		8	31	668	
6	29	611		8	31	736	
6	29	702		8	31	749	
6	29	736	500	8	31	806	706
6	30	525		8	32	430	
6	30	559		8	32	642	
6	30	571		8	32	702	
6	30	708	591	8	32	714	
6	31	413		8	32	747	
6	31	464		8	32	780	
6	31	584		8	32	793	687
6	31	612	518	8	33	718	
7	27	643	643	8	33	757	738
7	29	442		8	34	813	
7	29	467		8	35	493	
7	29	510		8	36	985	
7	29	536		9	29	590	
7	29	582		9	29	623	
7	29	611		9	29	792	668
7	29	781		9	31	611	
7	29	867	600	9	31	890	

TABLE 1.—*Static bending*—Continued.

Average thickness of wall.	Average outside circumference.	Crushing load.	Average crushing load for the same thickness of wall and the same outside circumference.	Average thickness of wall.	Average outside circumference.	Crushing load.	Average crushing load for the same thickness of wall and the same outside circumference.
mm.	*cm.*	*kg.*	*kg.*	*mm.*	*cm.*	*kg.*	*kg.*
9	31	1,020	840	9	35	917	
9	32	945		9	35	997	823
9	33	783		10	31	714	
9	33	797		10	31	815	765
9	33	856		10	32	928	
9	33	987		10	32	937	933
9	33	1,010	887	10	33	442	
9	34	702		10	34	1,392	
9	35	556		11	33	1,120	

TABLE 2.—*Static bending.*

[Span, 28 inches; beam loaded at center with the points of support and application of load at or near the nodes; speed of machine, 0.225 inch per minute.]

Average thickness.	Average outside circumference.	Crushing load.	Average crushing load for the same thickness of wall and the same outside circumference.	Average thickness.	Average outside circumference.	Crushing load.	Average crushing load for the same thickness of wall and the same outside circumference.
mm.	*cm.*	*kg.*	*kg.*	*mm.*	*cm.*	*kg.*	*kg.*
4	19	408		5	21	510	
4	21	447		5	21	513	
4	21	555	451	5	21	521	
4	22	426		5	21	558	
4	22	431		5	21	560	
4	22	434	445	5	21	567	
4	23	476		5	21	623	497
4	24	250		5	22	386	
4	24	424	337	5	22	397	
4	27	329		5	22	404	
5	18	497		5	22	424	
5	20	442		5	22	437	
5	20	562	502	5	22	475	
5	21	386		5	22	510	
5	21	422		5	22	525	
5	21	438		5	22	532	
5	21	442		5	22	533	
5	21	449		5	22	534	
5	21	467		5	22	600	
5	21	485		5	22	630	491
5	21	499		5	23	352	
5	21	499		5	23	386	
5	21	509		5	23	465	

TABLE 2.—*Static bending*—Continued.

Average thickness.	Average outside circumference.	Crushing load.	Average crushing load for the same thickness of wall and the same outside circumference.	Average thickness.	Average outside circumference.	Crushing load.	Average crushing load for the same thickness of wall and the same outside circumference.
mm.	*cm.*	*kg.*	*kg.*	*mm.*	*cm.*	*kg.*	*kg.*
5	23	475		5	26	445	
5	23	490		5	26	465	
5	23	491		5	26	479	
5	23	521		5	26	479	
5	23	522		5	26	480	
5	23	526		5	26	490	
5	23	535		5	26	495	
5	23	549		5	26	495	
5	23	600		5	25	522	
5	23	691	508	5	26	533	
5	24	352		5	26	545	
5	24	390		5	26	555	
5	24	394		5	26	558	
5	24	397		5	26	585	
5	24	397		5	26	600	
5	24	397		5	26	612	
5	24	422		5	26	635	
5	24	431		5	26	704	509
5	24	443		5	27	374	
5	24	447		5	27	522	
5	24	447		5	27	562	
5	24	465		5	27	580	510
5	24	465		6	21	454	
5	24	480		6	21	523	
5	24	485		6	21	586	554
5	24	493		6	23	493	
5	24	495		6	23	645	569
5	24	508		6	24	487	
5	24	521		6	24	554	
5	24	521		6	24	555	
5	24	533		6	24	612	
5	24	578		6	24	658	
5	24	615		6	24	658	
5	24	624		6	24	663	
5	24	625	482	6	24	667	607
5	25	388		6	25	513	
5	25	438		6	25	554	
5	25	442		6	25	624	564
5	25	462		6	26	397	
5	25	535		6	26	437	
5	25	612		6	26	521	
5	25	662	506	6	26	522	
5	26	397		6	26	525	
5	26	408		6	26	533	
5	26	430		6	26	534	
5	26	431		6	26	555	
5	26	435		6	26	562	
5	26	442		6	26	585	

TABLE 2.—*Static bending*—Continued.

Average thickness.	Average outside circumference.	Crushing load.	Average crushing load for the same thickness of wall and the same outside circumference.	Average thickness.	Average outside circumference.	Crushing load.	Average crushing load for the same thickness of wall and the same outside circumference.
mm.	*cm.*	*kg.*	*kg.*	*mm.*	*cm.*	*kg.*	*kg.*
6	26	588	----------	6	26	783	577
6	26	596	----------	6	27	422	----------
6	26	623	----------	6	27	510	----------
6	26	645	----------	6	27	534	489
6	26	646	----------	7	26	579	----------
6	26	658	----------	7	26	683	631
6	26	670	----------				

TABLE 3.—*Summary of static bending tests.*

Span, 60 inches.			Span, 28 inches.		
Average outside circumference.	Average thickness of walls.	Crushing load.	Average outside circumference.	Average thickness of walls.	Crushing load.
cm.	*mm.*	*kg.*	*cm.*	*mm.*	*kg.*
29	5	Max. 611 Ave. 424 Min. 351	23	4	Max. 555 Ave. 418 Min. 250
29	6	Max. 736 Ave. 510 Min. 283	24	5	Max. 704 Ave. 498 Min. 352
31	7	Max. 900 Ave. 639 Min. 442	25	6	Max. 783 Ave. 573 Min. 422
32	8	Max. 985 Ave. 687 Min. 430			
32	9	Max. 1,020 Ave. 817 Min. 556			
32	10	Max. 1,320 Ave. 886 Min. 442			

TABLE 4.—*Compression parallel to the grain.*

[Column test, height 48 inches; upper end of specimen U-shaped to take a rounded bearing surface; speed of machine, 0.042 inch per minute.]

Average thickness.	Average outside circumference.	Crushing load.	Average crushing load for the same thickness of wall and the same outside circumference.	Average thickness.	Average outside circumference.	Crushing load.	Average crushing load for the same thickness of wall and the same outside circumference.
mm.	*cm.*	*kg.*	*kg.*	*mm.*	*cm.*	*kg.*	*kg.*
8	30	6,730	----------	10	33	10,000	8,590
8	33	5,610	----------	10	34	8,450	----------
9	31	8,200	----------	10	35	7,100	----------
9	31	12,300	10,250	10	35	7,550	----------
9	32	5,970	----------	10	35	9,300	----------
9	32	7,230	----------	10	35	9,660	8,402
9	32	10,700	7,967	10	36	8,340	----------
9	33	7,160	----------	10	36	9,550	8,945
9	33	8,000	----------	10	38	9,610	----------
9	33	9,750	----------	11	33	9,000	----------
9	33	9,800	8,678	11	33	9,540	9,270
9	34	7,700	----------	11	34	4,630	----------
9	34	8,400	8,050	11	34	8,840	6,735
9	35	7,150	----------	11	35	8,000	----------
9	35	8,230	7,690	11	35	9,980	----------
9	36	6,330	----------	11	35	10,100	9,360
10	30	7,350	----------	11	37	9,240	----------
10	30	9,650	8,500	11	38	8,150	----------
10	31	6,420	----------	12	30	9,400	----------
10	31	9,470	7,945	12	31	6,040	----------
10	32	7,580	----------	12	31	7,600	6,820
10	32	7,940	7,760	12	35	9,650	----------
10	33	5,800	----------	13	29	10,400	----------
10	33	8,540	----------	13	32	10,250	----------
10	33	9,030	----------	13	34	10,200	----------
10	33	9,580	----------	13	38	11,900	----------

TABLE 5.—*Compression parallel to the grain.*

[Height, 14 inches; both ends cut at right angles to the axis; speed of machine, 0.042 inch per minute.]

Average thickness.	Average outside circumference.	Crushing load.	Average crushing load for the same thickness of wall and the same outside circumference.	Average thickness.	Average outside circumference.	Crushing load.	Average crushing load for the same thickness of wall and the same outside circumference.
mm.	*cm.*	*kg.*	*kg.*	*mm.*	*cm.*	*kg.*	*kg.*
4	17	3,660	----------	4	19	4,180	----------
4	17	5,080	4,370	4	19	4,260	----------
4	18	4,030	----------	4	19	4,540	----------
4	18	4,590	----------	4	19	5,520	4,675
4	18	5,100	----------	4	21	6,150	----------
4	18	8,470	555	4	21	6,340	6,245

TABLE 5.—*Compression parallel to the grain*—Continued.

Average thickness.	Average outside circumference.	Crushing load.	Average crushing load for the same thickness of wall and the same outside circumference.	Average thickness.	Average outside circumference.	Crushing load.	Average crushing load for the same thickness of wall and the same outside circumference.
mm.	*cm.*	*kg.*	*kg.*	*mm.*	*cm.*	*kg.*	*kg.*
4	22	6,470	----------	5	19	6,360	----------
5	14	3,050	----------	5	19	6,640	5,689
5	15	3,460	----------	5	20	3,840	----------
5	15	3,820	----------	5	20	4,050	----------
5	15	4,740	4,067	5	20	4,790	----------
5	16	4,250	----------	5	20	6,600	----------
5	17	4,670	----------	5	20	7,360	5,334
5	17	5,210	----------	5	21	4,140	----------
5	17	5,450	5,066	5	21	4,430	----------
5	18	4,700	----------	5	21	4,660	----------
5	18	5,080	----------	5	21	4,780	----------
5	18	5,450	----------	5	21	5,670	----------
5	18	5,600	----------	5	21	6,550	5,039
5	18	5,880	5,342	5	22	4,840	----------
5	19	4,850	----------	5	22	5,290	----------
5	19	5,060	----------	5	22	6,280	5,470
5	19	5,200	----------	5	24	4,840	----------
5	19	5,500	----------	5	24	7,950	6,395
5	19	5,630	----------	6	23	8,470	----------
5	19	5,650	----------	7	19	5,100	----------
5	19	6,310	----------				

TABLE 6.—*Summary of compression tests parallel to the longitudinal axis.*

Column tests.			Short strut tests.		
Average outside circumference.	Average thickness of walls.	Crushing load.	Average outside circumference.	Average thickness of walls.	Crushing load.
cm.	*mm.*	*kg.*	*cm.*	*mm.*	*kg.*
33	9	Max. 12,300 Ave. 8,350 Min. 5,970	19	4	Max. 8,470 Ave. 5,260 Min. 3,660
33	10	Max. 10,000 Ave. 8,470 Min. 5,800	19	5	Max. 7,950 Ave. 5,230 Min. 3,050
35	11	Max. 10,100 Ave. 8,610 Min. 4,630			
32	12	Max. 9,650 Ave. 8,170 Min. 6,040			
33	13	Max. 11,900 Ave. 10,690 Min. 10,200			

TABLE 7.—*Crushing strength in compression parallel to the grain and fiber stress in bending at the instant of failure of split bamboo.*

Compression parallel to the grain-crushing strength. Each value is an average of four tests.	Greatest fiber stress in bending at the instant of failure. Each value is an average of two tests.		Compression parallel to the grain-crushing strength. Each value is an average of four tests.	Greatest fiber stress in bending at the instant of failure. Each value is an averag of two tests.	
	Exterior side up.	Exterior side down.		Exterior side up.	Exterior side down.
kg. per cm².	*kg. per cm²*.	*kg. per cm²*.	*kg. per cm²*.	*kg. per cm²*.	*kg. per cm²*.
[a] 749	1,270	957	626	1,470	1,240
455	1,470	1,080	484	885	[b] 689
505	1,120	957	13,484	1,660	1,380
565	1,380	985	----------	1,760	1,600
624	1,590	957	----------	1,190	1,030
368	1,430	1,280	----------	1,590	1,260
512	1,550	1,100	----------	1,410	1,010
[b] 352	1,590	985	----------	1,350	1,240
459	[a] 2,160	1,570	----------	1,340	1,190
696	885	660	----------	1,260	1,030
482	1,350	997	----------	1,410	1,360
534	1,420	997	----------	1,560	1,290
357	1,320	1,150	----------	1,360	1,240
584	1,190	1,050	----------	1,490	1,230
514	1,710	1,100	----------	1,440	1,160
586	1,660	1,310	----------	1,730	1,290
610	2,010	[a] 1,830	----------	1,290	864
566	1,330	843	----------	1,630	1,440
542	1,190	983	----------	1,520	1,240
576	1,120	843	----------	*1,310*	*1,040*
524	1,050	975	----------	61,265	48,769
565	1,840	1,480	Average 535	1,425	1,134
649	975	857			

[a] Maximum. [b] Minimum.

TABLE 8.—*Split bamboo. Static bending and compression parallel to the grain.*

GREATEST FIBER STRESS AT THE INSTANT OF FAILURE.

		kg. per cm².
Static bending:		
With load applied on the hard exterior side	Maximum	2,160
	Average	1,425
	Minimum	885
With load applied on the soft interior side	Maximum	1,830
	Average	1,134
	Minimum	689

CRUSHING STRENGTH.

Compression parallel to the grain, height 1 inch	Maximum	749
	Average	535
	Minimum	352

Tables 7 and 8 show the average results of tests on split bamboo in bending and compression. Moisture determinations on these split bamboo specimens average 13 per cent computed on the dry basis. The fiber stress values are about three times as high as those found in red lauan, or Philippine mahogany, while the crushing strength in compression parallel to the grain is about one and a half times as much.

CONCLUSIONS

It can safely be concluded that for all engineering purposes a piece of bamboo about 30 centimeters in circumference when loaded at the center on a span of 5 feet can support 0.5 ton, and a piece of the same size when used as a post or column about 4 feet high can support 4 tons. The thicker the specimen, the stronger it is. Shorter spans and shorter posts will in general support greater loads although this relation may not be in exact mathematical proportion. When used as flooring the native practice of having the exterior side up is explained by its strength, rigidity, and hardness. In the case of wattling, split bamboo is woven so that some of the pieces are with the exterior side up and others with the exterior side down and the finished product is possessed both of strength and toughness.

ILLUSTRATIONS

PLATE 1. Bamboo beam test.
2. Bamboo column test.
3. Cross section of bamboo.

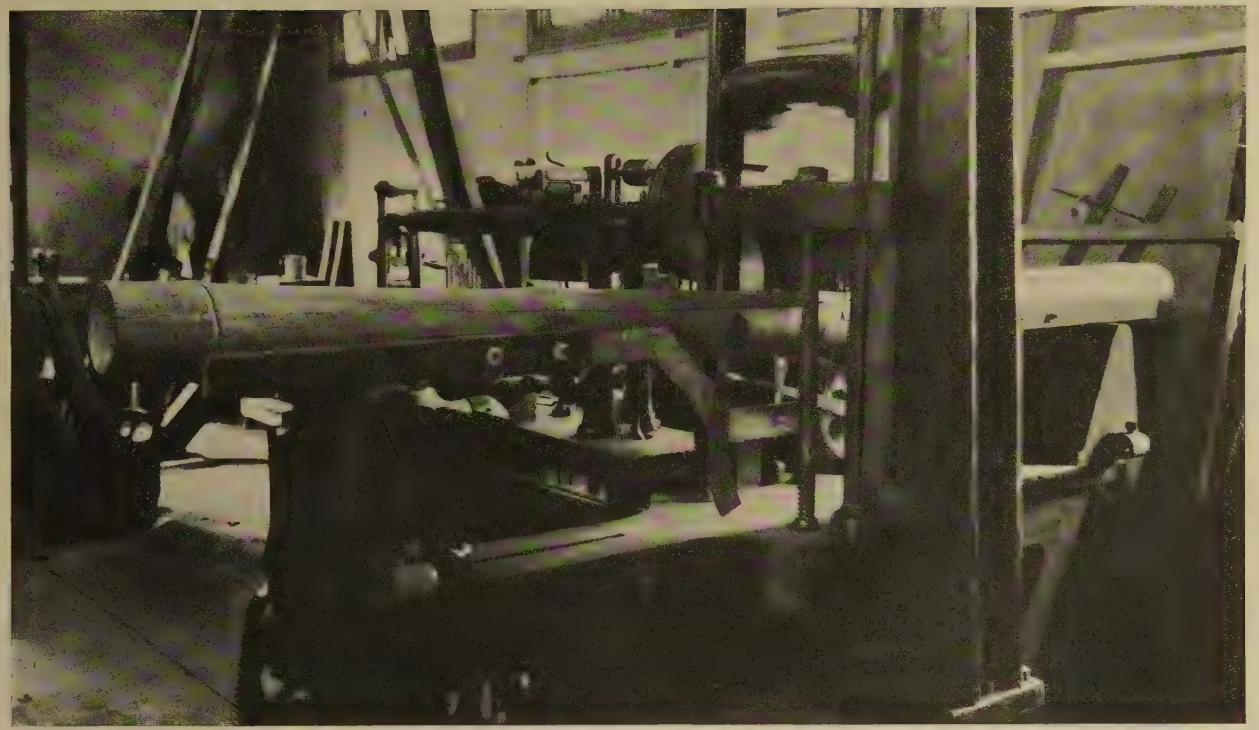

PLATE 1. BAMBOO BEAM TEST.

PLATE 2. BAMBOO COLUMN TEST.

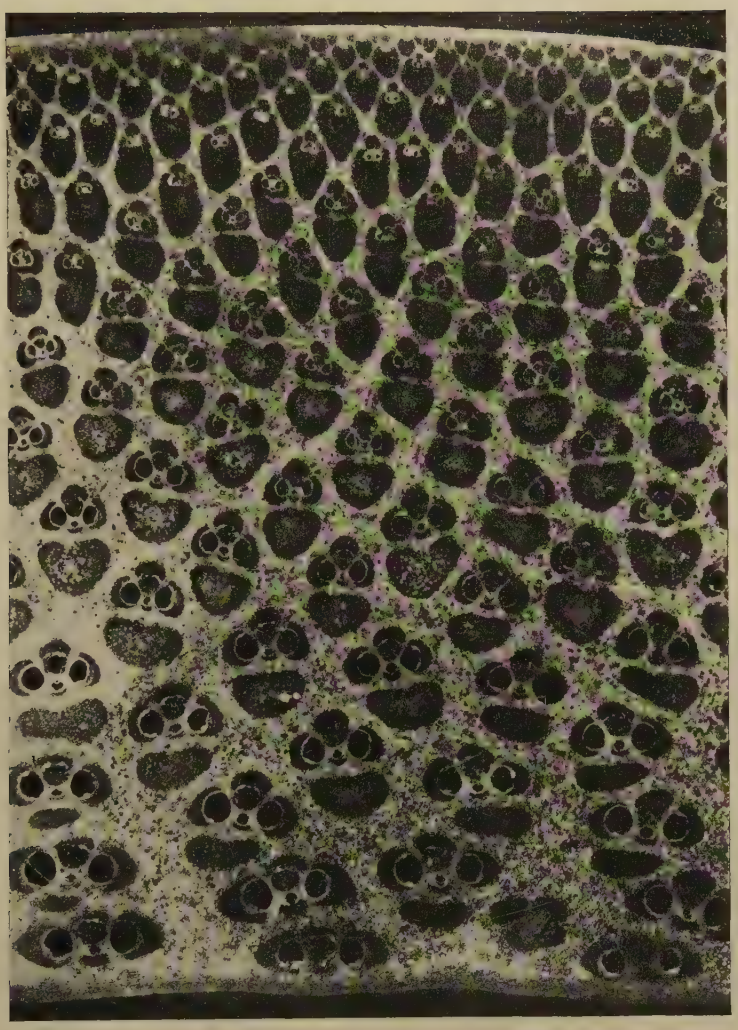

PLATE 3. CROSS SECTION OF BAMBOO.

ANTAMOKITE, A NEW GOLD-SILVER TELLURIDE

By A. D. Alvir
Of the Bureau of Science, Manila

ONE PLATE

A sample of gold ore from the Benguet Consolidated Mining Company mine at Antamok, Mountain Province, Philippine Islands, reported to run as high as 20,000 dollars a ton, was submitted to the Bureau of Science for study. The writer visited the mine and obtained more samples. This particular high-grade pocket has been called the Dickson vein, although it forms part of the 153-W-vein in the E level of the Antamok mine. This rich pocket averages about 2.5 meters in thickness and shows a splendid crustification of quartz and chalcopyrite with tetrahedrite. The country rock is andesite. A short distance from this pocket on the same vein some gypsum spar was observed in the middle of the vein, although in the rich pocket none was seen. In fact, in this level and in the lower ones gypsum is common.

Megascopically the ore is a fissure filling in andesite of more or less vuggy quartz impregnated with chalcopyrite and tetrahedrite which occur either alone or as a mixture. Pyrite is also present in small quantities, generally associated with the tetrahedrite.

Several sections were polished and studied under the microscope and camera-lucida sketches were prepared. In the course of this examination, a new gold-telluride mineral was observed.

Considerable trouble was met in the identification of this mineral because its microchemical reactions were very different from any known mineral whose microchemical reactions have been listed. These reactions with the reagents recommended by Davy and Farnham [1] are as follows:

HNO_3: Slowly tarnishes slightly dark and rubs nearly clean. Upon long interaction pits develop which persist even after a little re-grinding and re-polishing.

HCl: Negative (may tarnish very slightly).

[1] Microscopic Examination of the Ore Minerals, 1st ed., McGraw-Hill Book Co. (1920) 11.

KCN: Negative.
$FeCl_3$: Immediately tarnishes very iridescent and rubs same.
$HgCl_2$: Slowly tarnishes yellow and rubs faint yellow.
(KOH)H: Negative.

The hardness is low, probably between 2 and 3. The color under the microscope is grayish white with a slight bluish tinge. The color of the powder is dark gray.

The mineral occurs in very tiny portions almost always associated with the calaverite in the ore. Consequently it was almost impossible to isolate it for chemical analysis. However, after a long search, slightly larger portions of it were found, essentially free from calaverite. These were gouged out with a needle, and analyzed. There was not enough for a thorough chemical analysis, but what there was showed gold, tellurium, and traces of silver. No lead, antimony, or copper was found, showing that in all probability it is a gold telluride with a little silver. This mineral, quite conclusively a new species, is hereby named antamokite, after Antamok.

Under the microscope, the following minerals were identified given in the order of deposition: Prismatic quartz crystals; pyrite (in minor amounts); tetrahedrite (cut by calaverite); calaverite (silver free); antamokite; tetrahedrite (inclosing calaverite and antamokite); chalcopyrite (generally replacing tetrahedrite); and quartz filling.

Quartz in perfect prismatic crystals embedded in some of the ore minerals was the first to be deposited. The little pyrite found and the tetrahedrite come next, although some tetrahedrite certainly was deposited later. This tetrahedrite is auriferous but silver-free. Then the calaverite and the antamokite were deposited contemporaneously, generally in association with the tetrahedrite. Some portions show the two tellurides cutting the tetrahedrite, while other portions show that the tetrahedrite was deposited around the two tellurides. It seems that the tetrahedrite was deposited at two different stages. While some of the chalcopyrite may be primary, it occurs mostly as a replacement of the tetrahedrite. Finally more quartz was deposited. Tetrahedrite and chalcopyrite are the most abundant metallic minerals.

The deposit probably belongs to that class of young gold-silver lodes to which the Cripple Creek [2] deposits belong. They are generally quartzose fissure fillings, full of vugs, with tellu-

[2] Lindgren, Waldemar, Mineral Deposits, 2d ed., McGraw-Hill Book Co. (1919) 521–526.

rides of gold and silver, tetrahedrite, and the common sulphides as ore minerals. These were precipitated from hot ascending solutions related to the latest instrusions. The country rock is generally andesite.

Ores of this class generally do not persist to depths greater than 500 or 700 meters. It has been observed that there is an impoverishment as the zone of primary minerals is attained. It is also known, as at Cripple Creek,[3] that there are no signs of secondary enrichment. In the zone of oxidation metallic gold should be found derived from the oxidation of the tellurium in the tellurides.

This ore carries much gold, very little silver, and a considerable amount of copper.

[3] Lindgren, op. cit. 474 and 883.

ILLUSTRATION

PLATE 1. Antamokite; *a*, antamokite; *c*, calaverite; *qtz*, quartz.

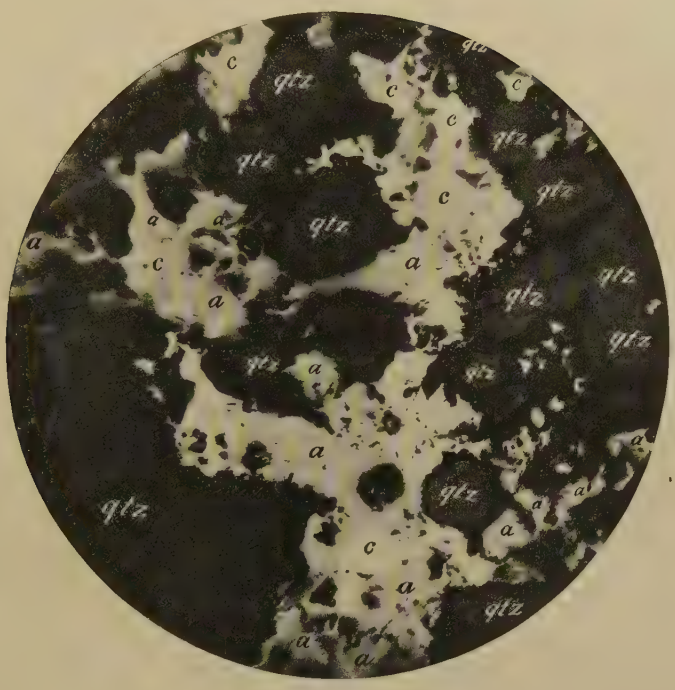

PLATE 1. ANTAMOKITE.

COPEPODA HARPACTICOIDA VON DER INSEL LUZON, PHILIPPINEN

Von P. A. CHAPPUIS
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MIT EINER FIGUR

Im Sommer 1926 hatte der leider zu früh dahingeschiedene Prof. C. F. Baker vom College of Agriculture in Los Baños die Freundlichkeit mir einige Planktonproben aus der grossen Laguna de Bay in Central Luzon zu senden. Diese Proben wurden am 8.V.26. von einem Schiffchen aus an einer stark mit *Myriophyllum* bewachsenen Stelle gesammelt, und in Alkohol konserviert. Die Untersuchung ergab, dass sie neben zahlreichen Cladoceren, grosse Mengen von Centropagiden und Cyclopiden, einige Harpacticiden sowie auch Hydracarinen enthielt.

Die Cladoceren verblieben leider unbestimmt, die anderen Gruppen jedoch wurden von Spezialisten bearbeitet deren Resultate in dieser und in zwei folgenden Arbeiten verüffentlicht werden sollen. Dieses Material war uns sehr wertvoll, umsomehr als aus den Philippinen bis heute sozusagen nichts bekannt war.

In den drei Proben aus der Laguna de Bay, central Luzon, fanden sich nur drei verschiedene Harpacticidenarten vor, und zwar: ein leider noch nicht geschlechtsreifes und desshalb unbestimmbares Exemplar das, soweit ersichtlich, einer marinen oder Brackwasserform nahestehen muss; drei Weibchen von *Nitocra platypus bakeri* subsp. nov.; und zwei Weibchen von *Canthocamptus bidens* subsp. *coronatus* (Sars).

NITOCRA PLATYPUS BAKERI subsp. nov.

Weibchen.—Körper schlank, die ersten Antennen achtgliedrig; Sinneskolben des vierten Gliedes erreicht das Ende der Antenne nicht. Nebenast der zweiten Antenne dreieckig, mit drei distalen Borsten. Rostrum schmal, fast so lang wie das erste Glied der ersten Antenne. Sämmtliche Thoracalsegmente ohne Dornenreihen, aber mit zahlreichen feinen Sinnesborsten.

Am ersten Abdominalsegment, ventral, in der Mitte, eine Reihe sehr feiner Dornen; nahe dem Hinterrande eine zweite

Reihe gröberer Dornen. Am zweiten und dritten Segment findet sich eine ähnliche ventrale Dornenreihe; am vierten Segment ist die Dornenreihe in der Mitte der Segmenthöhe. Ausserdem ist die Basis der Furkaläste ventral mit Zähnchen umsäumt; auf der Dorsalseite finden sich jederseits zwei Dornen.

Analoperculum mit vier bis fünf groben Dornen. Furka wenig länger wie breit, mit zwei wohl entwickelten apicalen Borsten von denen die innere mehr wie doppelt so lang ist wie die äussere. Die geknöpfte Borste subapical, ein bischen auf die Innenseite gerückt.

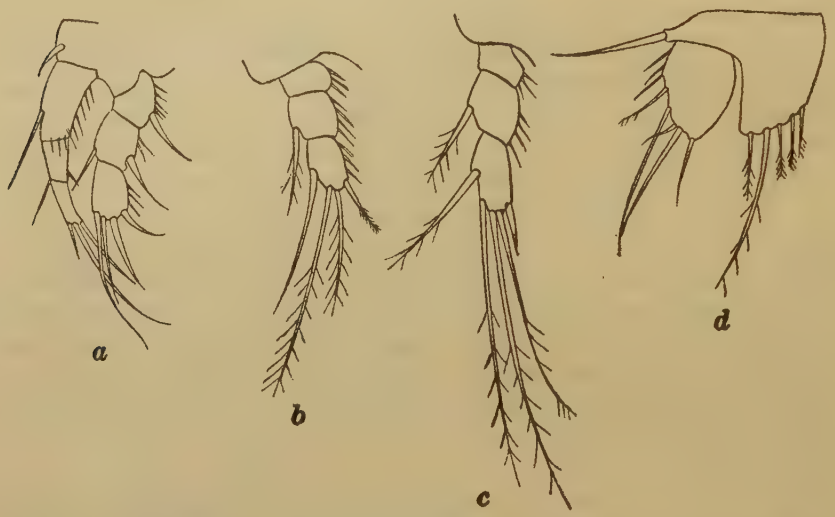

FIG. 1. *Nitocra platypus bakeri* subsp. nov. Weibchen, *a*, Erstes Beinpaar; *b*, Endopodit des zweiten Beinpaares; *c*, Endopodit des dritten Beinpaares; *d*, fünftes Beinpaar.

Endopodit des ersten Beinpaares (Fig. 1, *a*) gleich lang wie der Exopodit; sein letztes Glied apical mit einem Dorn, einer langen und einer kurzen Borste bewehrt. Letztes Glied des Exopoditen mit nur zwei Dornen und zwei Borsten. Erstes Glied des Endopoditen P_2 (Fig. 1, *b*) ohne Innenranddorn, zweites Glied mit Innenrandborste, drittes Glied mit drei Borsten und einem Dorn. Endopodite des P_3 und P_4 (Fig. 1, *c*) gleich gebaut: Erstes Glied ohne Innenranddorn, zweites Glied mit Innenrandborste, drittes Glied mit vier Borsten und einem Dorn. Die Exopodite dieser drei Beinpaare sind ähnlich gebaut: das erste Glied mit einem Dorn an der äusseren distalen Ecke; zweites Glied mit einem Dorn an der gleichen Stelle und einer Borste

an der distalen Ecke der Innenseite; letztes Glied mit drei Dornen am Aussenrand, zwei apicalen Borsten und zwei Borsten am Innenrande. Von diesen zwei Borsten ist die proximale beim P_2 und P_3 nicht sehr stark entwickelt.

Das Basalglied des P_5 (Fig. 1, *d*) breit; der innere Teil ist vorgezogen, erreicht das Ende des zweiten Gliedes und trägt fünf Borsten von denen die vierte, von Innen gezählt, die grösste ist. Die anderen sind untereinander ungefähr gleich lang. An der äusseren distalen Ecke, neben der letzten Borste, finden sich zwei kleine Dörnchen. Das zweite Glied ist breit, oval, mit fünf grösseren und einigen kleineren Borsten.

Das Männchen ist unbekannt.

Fundort, Laguna de Bay, Central Luzon. Drei Weibchen mit Eiballen.

Diese Art ist mit *Nitocra platypus* Daday aus einem Tümpel bei Wat-Sabatomo, Siam,[1] so nahe verwandt, dass sie als Unterart derselben betrachtet werden muss. Die Hauptunterschiede zwischen der Unterart und *N. platypus* sind forgende:

P_1

N. platypus, typ., Exopodit, drittes Glied, mit 2 Borsten und 3 Dornen.
N. platypus bakeri, Exopodit, drittes Glied mit 2 Borsten und 2 Dornen.

P_4

N. platypus typ., Endopodit, erstes Glied mit Innenranddorn.
N. platypus bakeri, Endopodit, erstes Glied ohne Innenranddorn.

CANTHOCAMPTUS BIDENS subsp. CORONATUS (Sars).

Attheyella coronata Sars 1904.
Canthocamptus bidens Daday 1905.
Attheyella decorata Daday 1907.
Canthocamptus bidens van Douwe 1912.

Vergleichen wir Punkt für Punkt Schmeil's Beschreibung von *C. bidens* und diejenige Sars's für *Attheyella coronata*, so ist ausser der Zweigliedrigkeit des Endopoditen P_1 kein wesentlicher Unterschied festzustellen. Dies veranlasste van Douwe[2] die Vermutung auszusprechen "dass dem sonst so vorsichtigen Schmeil hier tatsächlich ein Versehen unterlaufen ist und er die Trennung der beiden letzten Innenastglieder übersehen hat,

[1] Zool. Jahrb. Abt. Syst. 24 (1907) 175–206.
[2] Arch. f. Hydrob. 7: 316.

was insoferne wohl entschuldbar wäre, als Schmeil nur zwei Tiere bei Aufstellung der Art zur Verfügung hatte und die Trennungsstelle der beiden fraglichen Fussglieder nicht immer leicht zu erkennen ist."

Canthocamptus bidens wurde in Europa jedoch nicht nur von Schmeil gefunden, sondern auch von Scourfield und von Jakubisiak.[3] Letzterer bemerkt ausdrücklich, dass das von ihm gefundene Tier mit dem von Schmeil beschriebenen *C. bidens* vollständig identisch ist, und dass *A. coronata* hauptsächlichst durch die Dreigliedrigkeit des Endopoditen P_1 von dieser Art verschieden ist.

Auf eine briefliche Anfrage von mir, antwortete Herr Jakubisiak: "Je puis vous affirmer que l'exemplaire de *C. bidens*, trouvé par moi, avait l'endopodite de la première paire de pattes natatoire composé de deux articles et non de trois."

Es scheint also festzustehen das Schmeil sich nicht geirrt hat, und dass eine *Canthocamptus* Art wie er sie unter dem Namen *C. bidens* beschrieben hat tatsächlich existiert. Die Reduktion einer Gliedmasse ist ja bei Harpacticiden keine Seltenheit, und wir haben es hier mit einer ähnlichen Erscheinung zu tun wie bei der almählichen Reduktion der Gliederzahl der Endopoditen bei den Verwandten des *Canthocamptus minutus* Claus. Dort kann ja die fortschreitende Verringerung der Gliederzahl von *C. minutus* über *C. mrazeki* zu *C. Zschokkei* am besten beobachtet werden.

Die Reduktion einer Gliedmasse eines einzigen Beinpaares ist aber, wenn sie allein auftritt von so untergeordneter Bedeutung, dass wir sie nicht als Artmerkmal betrachten können. *Canthocamptus coronatus* muss also in der Nomenklatur als Subspecies von *C. bidens* figurieren, obwohl er morphologisch ursprünglicher und desshalb als Stammform von *C. bidens* angesehen werden muss.

Zu den Synonyma ist folgendes zu bemerken: Über verschiedene von Daday beschriehene *Canthocamptus* Arten ist schon in einer früheren Arbeit berichtet worden[4] und es konnte bei dieser Gelegenheit festgestellt werden, dass den Daday'schen Beschreibungen, wenigstens was die Harpacticiden betrifft, nicht ohne weiteres zu trauen ist. Ein typisches Beispiel von Daday's Arbeitsweise giebt uns seine Beschreibung von *Attheye-*

[3] Bull. Soc. Zool. France 47.

[4] Chappuis, P. A., Bull. Soc. Sc. Cluj 2 (1924) 96–103.

lla decorata.[5] Dort sagt er: "Ich habe diese Art zuerst aus Neuguinea beschrieben, allein bei der Vergleichung mit neuerem mir vorgelegenen Material stellte es sich heraus, dass das Exemplar aus Neuguinea ein junges, noch nicht geschlechtsreifes Tier war .....Aus der Fauna von Paraguay habe ich dieselbe als Varietät von *Canthocamptus bidens* Schmeil beschrieben, allein dort habe ich bloss geschlechtsreife Exemplare erhalten, war somit nicht in der Lage, die Identität mit dem neuguinesischen *Canthocamptus decoratus* zu constatieren."

Canthocamptus bidens coronatus ist nun sehr nahe mit *C. grandidieri* Richard und vielen anderen tropischen *Canthocamptus* Arten verwandt, und es ist unmöglich zu entscheiden ob die Daday vorgelegene Jugendform aus Neu-Guinea wirklich zu *C. coronatus* gehört. Bevor jedoch Daday merkte, dass der als *C. decoratus* beschriebene Harpacticide nur ein Jugendstadium einer noch unbekannten Art war, beschrieb Sars seine *Attheyella coronata* aus den Hawaiischen Inseln, und, da diese Beschreibung die erste ist die sich auf ein geschlechtsreifes Tier dieser Species bezieht, so müssen wir der Bezeichunng von Sars die Priorität geben.

Die Verbreitung dieses Tieres, ist der von *C. grandidieri* ähnlich, nur ist unser Subspecies bis jetzt noch nicht aus Afrika bekannt; wohl aber aus den Hawaii Inseln (Sars); Paraguay (Daday); Sumatra und Java (Daday); Ceylon (nach einer brieflichen Mitteilung von F. Kiefer der diese Form in Material aus dem Gregory-See fand); Brasilien (van Douwe); und nun aus den Philippinen.

Systematisch gehört *C. bidens coronatus* einer in den Tropen weit verbreiteten Harpacticidengruppe an für welche V. Brehm die Schaffung einer besonderen Gattung *"Chappuisiella"* vorschlägt.[6] Ueber die Diagnose und Zusammensetzung dieser Gruppe wird in einer späteren Arbeit die Rede sein.

[5] Zool. Jahrb. 24 (1907).
[6] Arch. f. Hydrob. 16.

ERKLÄRUNG DER FIGUR

FIG. 1. *Nitocra platypus bakeri* subsp. nov. Weibchen, *a*, Erstes Beinpaar; *b*, Endopodit des zweiten Beinpaares; *c*, Endopodit des dritten Beinpaares; *d*, fünftes Beinpaar.

SÜSSWASSER COPEPODEN (CALANOIDA UND CYCLOPOIDA) VON DER INSEL LUZON, PHILIPPINEN

Von FRIEDRICH KIEFER
Dilsberg (bei Heidelberg)

MIT EINER FIGUR

Herr Dr. P. A. Chappuis[1] war so liebenswürdig, mir vor einiger Zeit drei Gläschen mit Planktonmaterial aus der Laguna de Bay von der Insel Luzon zu senden, damit ich die darin vorkommenden Copepoda-Calanoida und -Cyclopoida untersuche Es sei mir zunächst gestattet, Herrn Dr. Chappuis für Ueberlassung dieser Proben auch an dieser Stelle meinen herzlichsten Dank auszusprechen. Eine über Süsswasser-Copepoden der Philippinen handelnde Arbeit ist mir bis jetzt noch nicht bekannt geworden. Ich ging darum mit einiger Spannung an die Bestimmung der gefundenen Tiere; und obwohl die Proben sehr klein waren, konnte ich doch 4 verschiedene Arten aus den beiden oben genannten Unterordnungen feststellen.

CALANOIDA

DIAPTOMUS SENSIBILIS sp. nov. Fig. 1.

Von den Calanoida fand sich nur eine Art und zwar nur in wenigen Stücken. Es ist ein *Diaptomus*, den ich mit keiner mir bekannten Art identifizieren konnte. Er sei deshalb unter obigem Namen in die Wissenschaft eingeführt.

Das Weibchen.—Der Körper des ungefähr 1350 μ langen Tierchens ist ziemlich schlank. Grösste Breite des Vorderkörpers etwa in der Mitte. Viertes und fünftes Thoraxsegment sind dorsal vollkommen miteinander verwachsen, nur seitlich lässt sich noch die ehemalige Trennung erkennen (Fig. 1, *a*). Die Flügel des letzten Segments sind nur recht klein und gerade nach hinten gerichtet; am Ende sitzt jederseits ein schlanker, nahezu zylindrischer Sinnesdorn (Fig. 1, *a*). Das schlanke Abdomen ist dreigliedrig. Das Genitalsegment ist rund doppelt so lang wie die beiden andern zusammen und in seinem vor-

[1] Das Material wurde von Prof. C. F. Baker vom College of Agriculture in Los Baños, Philippine Islands, gesammelt und im Jahre 1926 Dr. P. A. Chappuis zugesandt.—EDITOR.

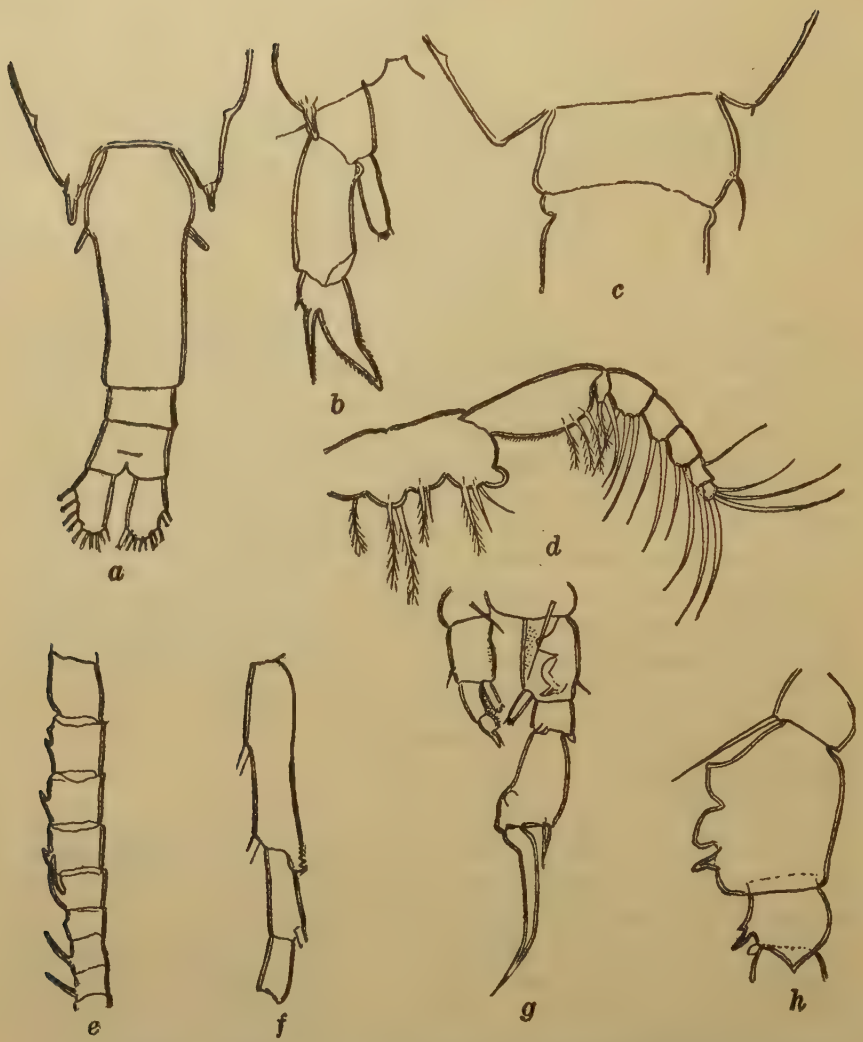

FIG. 1. *Diaptomus sensibilis* sp. nov.; *a*, viertes und fünftes Thoraxsegment und Abdomen, Weibchen, dorsal; *b*, rudimentärer Fuss des Weibchens; *c*, fünftes Thoraxsegment und Genitalsegment, Männchen, dorsal; *d*, grosser Maxilliped, Weibchen; *e*, mittlerer Teil der männlichen Greifantenne; *f*, Endglieder der männlichen Greifantenne; *g*, rudimentäres Beinpaar des Männchens; *h*, erstes und zweites Basalglied und erstes Exopoditglied des rechten männlichen Fusses von der Seite.

deren Teile jederseits mässig und annähernd symmetrisch verbreitert. Unterhalb der breitesten Stelle sitzt jederseits ebenfalls ein langer zylinderförmiger Sinnesdorn (Fig. 1, *a*). Die beiden übrigen Abdominalsegmente sind symmetrisch, das mittlere deutlich kürzer als das letzte. Lie beiden Furkaläste sind ebenfalls einander gleich und ohne erwähnenswerte Kennzeichen.

Die Vorderantennen reichen zurückgeschlagen etwas über das Hinterende der Furkaläste hinaus, sind also sehr lang. Bau und Bewehrung des grossen Maxillipeden sind am besten aus Fig. 1, *a*, zu erkennen. Besonders hingewiesen sei darauf, dass Lobus 3 des ersten Basalgliedes nur 2, Lobus 4 nur 3 Borsten besitzt. Ueber die Schwimmbeine ist hier nur zu sagen, dass am mittleren Gliede des Innenastes vom zweiten Paar jegliche Spur eines sogenannten Schmeilschen Anhanges fehlt. Das rudimentäre Füsschen ist folgendermassen gebaut: (Fig. 1, *b*). Das breite erste Basalglied trägt einen hyalinen Sinnesdorn, der denen an den Thoraxflügeln überaus ähnlich ist. Das zweite Basalglied, and dessen kurzem äusseren Rande die übliche Borste sitzt, ist abgestumpft kegelförmig. Das erste Aussenastglied, das rund doppelt so lang ist wie breit, zeigt an seiner proximalen Innenecke einen chitinigen Vorsprung. Das zweite Exopoditglied ist in eine nur mässig nach innen gekrümmte, beiderseits mit Dörnchenfiederchen versehene, starke Klaue ausgezogen. Das kleine dritte Glied ist vollkommen mit dem zweiten verschmolzen; es ist apikal mit einem starken Stachel und einer zärteren Borste bewehrt. Ausserdem sitzt an der Basis des ehemaligen Gliedes ein dem Aussenrand des zweiten Gliedes zugehörendes kleines Dörnchen. Der zylindrische, an seinem Ende kegelförmig zugespitzte Innenast erreicht etwa zwei Drittel bis drei Viertel der Länge des ersten Aussenastgliedes. Seine hyaline Spitze trägt einen Kranz feiner Börstchen und innen ein gröberes Stachelchen. Eiballen wurden keine beobachtet.

Das Männchen.—Es ist etwas kleiner und noch schlanker als das Weibchen. Viertes und fünftes Thoraxsegment sind ebenfalls miteinander verwachsen. Das Genitalsegment trägt rechts seitlich einen langen schlanken Sinnesdorn (Fig. 1, *c*). Das vorletzte Abdominalsegment ist hinten schräg abgeschnitten und rechts etwas mehr nach hinten gezogen als links. Ueber die symmetrischen Furkaläste ist hier nichts besonderes zu sagen. Die Greifantenne ist in ihrem mittleren Teil nur sehr schwach aufgetrieben. Die Glieder 10, 11, 13 bis 16 sind mit Dornen bewehrt; über deren Grösse und Aussehen unterrichtet am besten (Fig. 1, *e*). Das drittletzte Glied ist in einen Fortsatz ausgezogen, der ein Drittel bis ein Viertel der Länge des folgenden Gliedes erreicht. Von der Mitte des Gliedes ab ist sein Rand auf der Fortsatzseite von einer schmalen hyalinen Membran gesäumt, die unmerklich in die Zähnelung des Fortsatzes übergeht (Fig. 1, *f*). Das Endglied ist ohne besondere Bewehrung. Ueber die Mundwerkzeuge und die Schwimmbeine ist nichts

neues zu sagen. Das rudimentäre Fusspaar (Fig. 1, *g*) weist folgenden Bau auf:

Rechts.—Das kurze, aber recht breite erste Basalglied besitzt auf seiner kaudalen Fläche einen sehr langen, borstenförmigen Sinnesstachel. Das Zweite Basalglied, in Aufsicht etwa rechteckig und doppelt so lang wie breit, trägt auf seiner kaudalen Fläche drei Chitin-Auswüchse, deren Aussehen in Seitenlage des Fusses am besten die beigegebene (Fig. 1, *h*) zeigt. Die Innenseite des Gliedes weist eine feine Granulierung auf, die wohl als Sinnespolster gedeutet werden darf. Das erste Aussenastglied, das sehr kurz ist, besitzt zwei bemerkenswerte Fortsätze, einen auf der kaudalen Fläche sitzenden und nach innen gerichteten und einen an der distalen Aussenecke. Das zweite Exopoditglied verbreitert sich von der Basis zum distalen Ende zu deutlich. Ausser dem verhältnismässig kurzen, zweimal schwach geknickten Enddorn und dem ebenfalls nur recht kurzen, nahe der Basis der Klaue entspringenden Seitenranddorn, bemerkt man auf der Fläche des Gliedes in seiner unteren Hälfte und nahe dem Innenrande einen zarten hyalinen Auswuchs. Der zylindrische Innenast ist nur so lang wie das erste Aussenatglied.

Links.—Der ganze Fuss erreicht nur etwas mehr als die Länge des rechten bis zur Basis des ersten Exopoditgliedes, ist also ziemlich kurz. Das erste Basalglied besitzt einen ähnlichen, nur etwas kürzeren Sinnesdorn wie das entsprechende Glied der andern Seite, das zweite Glied ebenso ein Sinnespolster an seiner Innenseite. Der Aussenast erscheint zweigliedrig; er ist ziemlich einfach gebaut, trägt innen einige Sinnespolster, und das Ende des letzten Gliedes ist fingerförmig vorgezogen; neben diesem Fortsatz sitzt eine stachelförmige Borste. Der Innenast ähnelt dem des rechten Fusses in hohem Grade, nur ist er etwas kleiner (Fig. 1, *g*). Spermatophoren wurden nicht gesehen.

Hauptkennzeichen der Art sind: Form der weiblichen Thoraxflügel, die Sinnesdornen an diesen Flügeln und am Genitalsegment sowie die verschiedenen hyalinen Dornen und Höckerfortsätze am rudimentären männlichen Fusspaar.

CYCLOPOIDA

Im untersuchten Materiale fanden sich ausser dem eben beschriebenen *Diaptomus* noch Exemplare dreier verschiedener Cyclopoiden-Arten. Am häufigsten war *Mesocyclops leucharti* Cls., jedoch nicht in der in Europa vorkommenden Form, sondern in einer sich davon wohl unterscheidenden, in den Tropen

weit verbreiteten Variation, die vielleicht als Unterart angesehen werden kann; ich werde sie in einer demnächst erscheinenden Revision der Gattung *Mesocyclops* Sars genauer kennzeichnen.

Ihr gegenüber trat in den vorliegenden Proben eine zweite Art derselben Gattung zahlenmässig sehr stark zurück; nämlich, *Mesocyclops hyalinus* Rehbg.

Die dritte festgestellte Art endlich gehört in die *Cyclops varicans*-Gruppe. Ihre genaue systematische Stellung kann jedoch erst ermittelt werden, wenn die in Vorbereitung befindliche Bearbeitung der *varicans*-Aehnlichen abgeschlossen its. Ich verzichte daher an dieser Stelle auf weitere Ausführungen über die wenigen gefundenen Exemplare.

Aus dem spärlichen hier behandelten Material lassen sich natürlich weder in oekologischer noch in zoogeographischer Hinsicht irgendwelche Schlüsse ziehen. Die drei Cyclopiden sind (wenigstens als Typen) weit verbreitet; der *Diaptomus* aber ist noch neu und vorerst noch ohne ganz sicheren Anschluss an bereits bekannte Formen. Es wäre daher sehr zu begrüssen, wenn ein mit den Lebensgewohnheiten der freilebenden Copepoden etwas vertrauter Süsswasser-Zoologe bald weitere Aufsammlungen von den Philippinen zusammenbrächte. Aus deren Bearbeitung müsste sich dann wohl erkennen lassen, ob die Philippinen (vor allem hinsichtlich ihrer Calanoiden-Fauna) zum malayischen Gebiet oder aber zu Südost-China nähere Beziehungen haben.

ERKLÄRUNG DER FIGUR

FIG. 1. *Diaptomus sensibilis* sp. nov.; *a*, viertes und fünftes Thoraxsegment und Abdomen, Weibchen, dorsal; *b*, rudimentärer Fuss des Weibchens; *c*, fünftes Thoraxsegment und Genitalsegment, Männchen, dorsal; *d*, grosser Maxilliped, Weibchen; *e*, mittlerer Teil der männlichen Greifantenne; *f*, Endglieder der männlichen Greifantenne; *g*, rudimentäres Beinpaar des Männchens; *h*, erstes und zweites Basalglied und erstes Exopoditglied des rechten männlichen Fusses von der Seite.

HYDRACARINEN VON DER INSEL LUZON, PHILIPPINEN

Von C. Walter
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MIT DREI FIGUREN

Meines Wissens sind von den Philippinen noch keine Hydracarinen beschrieben worden. Die drei im Nachfolgenden charakterisierten Arten verdanke ich Herrn Dr. P. A. Chappuis.[1] Sie stammen von Luzon. Zwei derselben, *Limnesia bakeri* und *Neumania flagellata*, stellen für die Wissenschaft neue Formen dar. Die dritte ist auf die sumatranische Species *Neumania ambigua* Piersig zu beziehen.

LIMNESIA BAKERI sp. nov. Fig. 1.

Männchen.—Verwandtschaftlich steht diese Art mit der von Kœnike[2] von Nossi-Bé beschriebenen *Limnesia aspera* sehr nahe; denn die Genitalplatten weisen nicht nur 6, sondern 8 Näpfe auf.

Die Körperlänge beträgt beim Männchen 0.540 Millimeter, die Körperbreite 0.420. Umrisslinie oval; der Stirnrand ist zwischen den 30 μ langen, 105 μ voneinander entfernten antenniformen Borsten vorgewölbt.

Die 5 μ dicke Haut ist nicht wie bei der Vergleichsart mit Chitinspitzchen bedeckt, sondern feinliniert. Auf dem Hinterrücken liegt eine kleine rundliche Platte als Muskelansatzstelle. Die grossen Vorderlinsen der Augen liegen 105μ auseinander, sind schmal elliptisch und erreichen den Körperrand. Hintere Linsen rundlich. Augenpigment rotbraun.

Ventralwand des Maxillarorganes 100 μ lang, an der Ansatzstelle der Palpen 80 μ breit. Der bis an den Hinterrand der Ventralwand reichende Pharynx erweitert sich hinten bis auf 23 μ, bleibt also im ganzen recht schmal. Hinterrand des Maxillarorganes quer abgeschnitten, seitwärts in je einen sehr kurzen Fortsatz auslaufend. Die oberen Fortsätze sind degegen lang und kräftig. Jederseits der Mundöffnung findet sich wie bei

[1] Das Material wurde von Prof. C. F. Baker vom College of Agriculture in Los Baños, Philippine Islands, gesammelt und im Jahre 1926 Dr. P. A. Chappuis zugesandt.—Editor.

[2] Abh. Senckenberg. naturf. Ges. 21 (1898) 407–410, Taf. 25, Fig. 114, 117.

der Vergleichsart ein zapfenförmiger Vorsprung mit kurzer Borste. Mandibel 0.163 Millimeter lang.

Streckseitenlängen der einzelnen Palpenglieder: Erste, 18; zweite, 73; dritte, 41; vierte, 88; fünfte, 33 μ. Der Palpus ist, wenn auch im ganzen recht ähnlich gebaut, so doch kürzer und gedrungener als derjenige der Vergleichsart. Der Chitinhöcker der Beugeseite des zweiten Gliedes kürzer, sein Zahn leicht nach vorn gerichtet. Tasthöcker des vierten Gliedes dem distalen Gliedende näher. Alles übrige its aus Fig. 1, *a* zu ersehen.

Das Epimeralgebiet bedeckt fast die vordere Hälfte der Ventralseite (Fig. 1, *b*). Hister der Maxillarbucht treten die ersten Epimeren sehr nahe zusammen, ohne jedoch miteinander zu verwachsen. Der gemeinsame Fortsatz der ersten und zweiten Epimere steht nicht über den verdickten Hinterrand vor. Der Aussenrand der vierten Platte in seiner ganzen Länge auswärts vorgebogen.

Die Beborstung der Beine ist eine mässige; insbesondere fällt die Reduktion der Schwimmhaare auf. Solche treten vereinzelt auf dem fünften Gliede des dritten und auf dem vierten und fünften Gliede des letzten Beines auf. Die die Gliedenden einnehmenden Borsten sind relativ lang und zum Teil gefiedert. Endglied des Hinterbeines ohne Krallen, vor der Basis des Chitinstiftes eine verlängerte Borste, im übrigen drei bis vier kürzere Borsten tragend. Kralle der drei Vorderbeine aus einem starken Hauptzahn und je einem ausserordentlich feinen Innen- und Aussenzahn bestehend. Länge der Beine: Erstes, 0.372; zweites, 0.435; drittes, 0.455; viertes, 0.590 Millimeter.

Das 0.125 Millimeter lange und 0.120 Millimeter breite Genitalorgan ist breit-oval und verlängert sich an seinem Hinterrande nach hinten in einen flächigen Vorsprung. Die Genitalöffnung hat eine Länge von 57 μ, ist elliptisch und von vier Napfpaaren umstellt, von denen das vordere und das hintere grösser sind als die beiden innern. Die beiden innern Näpfe einer Plattenhälfte sind weiter voneinander entfernt als diese von den äussern. Zwischen den beiden Näpfen des hintern Paares liegen zwei Verdickungen der Platte, die als Muskelansatzstellen dienen. Auf der Platte finden sich wenige feine Haare.

Excretionsporus nahe am Hinterrand des Körpers gelegen.

Weibchen.—Eiertragende Weibchen werden bis 0.9 Millimeter lang. Sie tragen die meisten Merkmale der Männchen. Der Palpus ist schlanker. Die Streckseitenlängen seiner Glieder messen: Des ersten Gliedes, 26; des zweiten, 104; des dritten, 67; des vierten, 137; des fünften, 44 μ. Die Beugeseitenhöcker

am vierten Gliede treten schwächer hervor. Genitalorgan (Fig. 1, *c*) 0.187 Millimeter lang, 0.150 Millimeter breit. Es hat ovalen Umriss. Jede Klappe trägt vier Näpfe in ähnlicher Anordnung wie beim Männchen, der Abstand zwischen der vorderen und der hinteren Gruppe ist jedoch grösser. Der vordere und der hintere Napf sind grösser als die beiden mittleren. Der vordere Stützkörper dem Vorderrande der Klappen angeschmiegt.

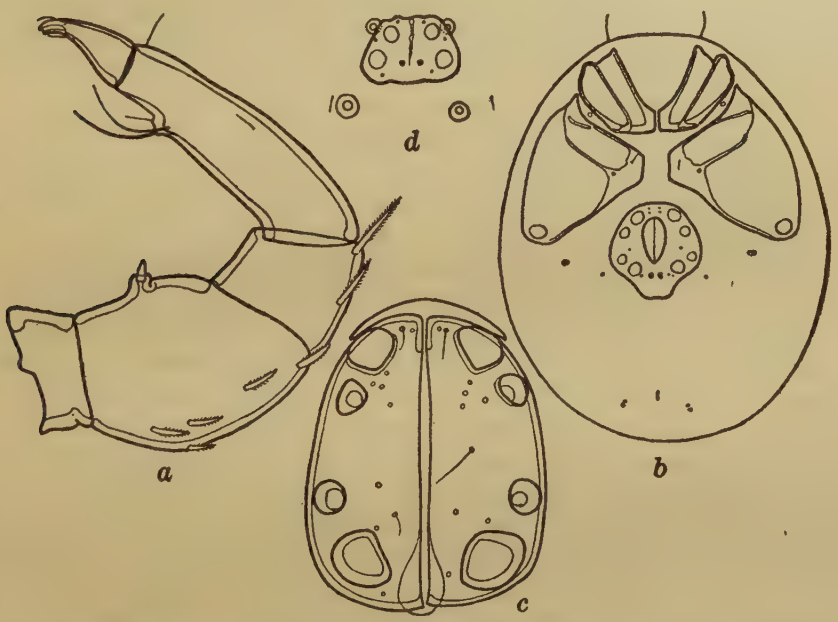

FIG. 1. *Limnesia bakeri* sp. nov.; *a*, Weibchen, Palpus; *b*, Männchen, Ventralansicht; *c*, Weibchen, Genitalorgan; *d*, Genitalplatte der Nymphe und die vier Näpfe des Teleiophanorganes.

Nymphe.—Die trapezartige Genitalplatte (Fig. 1, *d*) trägt zwei Paare von Näpfen, von denen das vordere kleiner ist als das hintere, welches vom Hinterrand weiter absteht als das vordere vom Vorderrand. Diese beiden Ränder leicht gewellt. Dem Palpus fehlt der Chitinhöcker auf der Beugeseite des zweiten Gliedes.

Teleiophanstadium.—Länge, 0.555 Millimeter. Teleioderma fein papillös. Teleiophanorgan 4-näpfig (Fig. 1, *d*), wodurch sich die 8-näpfigen *Limnesia*-Arten von den 6-näpfigen, deren Teleiophanorgan sich aus nur zwei Näpfen zusammensetzt, zu unterscheiden scheinen. Sollte diese Tatsache zur Regel wer-

den, so wäre dies ein wichtiger Grund zur Abtrennung der 8-näpfigen von den 6-näpfigen Limnesien.

Fundort.—Philippinen, Luzon. Ein Männchen, drei eiertragende Weibchen, ein Teleiophan-Stadium, eine Nymphe.

NEUMANIA AMBIGUA Piersig. Fig. 2.

Weibchen.—Von den Philippinen lag ein einziges weibliches Exemplar vor, das ich mit drei Weibchen dieser Art aus der Piersig'schen Sammlung vergleichen konnte. Piersig hat zu seiner Beschreibung [3] offenbar ein noch nicht völlig erwachsenes Exemplar verwendet, woraus sich manche Abweichung als bloss scheinbar bestehend herausstellt. Mit grosser Wahrscheinlichkeit dürfte sich auch *N. megalommata* Kœnike,[4] obwohl sie in ihren Körpermassen hinter denjenigen der Piersig'schen Individuen zurückbleibt, auf *N. ambigua* beziehen lassen. Ein abschliessendes Urteil ist vor Ueberprüfung der Type Koenike's nicht möglich.

Körperlänge des Philippinen-Exemplares 1.140 Millimeter, Haut weich, nicht zu Erhärtungen neigend, wie von Piersig vermutet. Epidermis auch bei den Vergleichstieren undeutlich liniert, mit zahlreichen, sehr feinen Chitinspitzchen dicht besetzt. Die meisten Drüsenmündungen höckerig erhaben, besonders stark zwei Drüsenöffnungen am Hinterrand des Körpers.

Maxillarorgan 130 μ lang, die Mandibeln nur wenig länger; deren Klauenglied wenig gebogen, 47 μ lang. Streckseitenlängen der Palpenglieder: Des ersten Gliedes, 31; des zweiten, 101; des dritten, 60; des vierten, 104; des fünften, 28 μ. Auf der Mitte der Flachseite gemessen entspricht die ermittelte Länge (0.265 Millimeter) genau dem von Koenike für *N. megalommata* angegebenen Palpenmasse. Für die Art ist die Einlenkung des Chitinzahnes in den Höcker an der distalen Beugeseite des vierten Gliedes charakteristisch: der Zahn sitzt nicht zentral, sondern in der Flanke des Höckers. Piersigs Fig. 28, Taf. 15, stellt den von der Aussenseite gezeichneten Palpus dar; es sind jedoch nicht alle Borsten eingezeichnet (siehe Fig. 2, *a*).

Bei eiertragenden Weibchen bedecken die Epimeren (Fig. 2, *b*) eine Fläche von 0.525 Millimeter Länge. Nach Piersig gleichen sie denen von *N. volzi.* Immerhin sind einige wesentliche Abweichungen zu vermerken: Hinterrand der vierten Epimere fast gerade, da die Einlenkungsstelle des Hinterbeines am Auss-

[3] Zool. Jahrb. Syst. 23 (1906) 329–330, Taf. 15, Fig. 28–30.

[4] Jahrb. Hamburger wiss, Anst., Mitt. naturh. Museum 23 (1906) 111–114, Taf. 1, Fig. 6–9.

enrande sehr weit nach hinten verlagert ist. Die Aussenfortsätze der dritten und der vierten Platte beide gerundet, bei *N. volzi* dagegen derjenige der dritten Epimere eckig, derjenige der vierten in einen schief rück- und auswärts gerichteten Zahn auslaufend. Der Hinterrandsfortsatz der vierten Epimere ist nur nach erfolgter Präparation erkennbar, da er ausserordentlich schwach chitinisiert ist. Dritte Epimere halb so lang wie die vierte.

Die Beine messen: Erstes, 0.945; zweites, 0.990; drittes, 0.945; viertes, 1.110 Millimeter. Wenn Piersig die Gliedmassen von *N. ambigua* als ähnlich ausgerüstet wie bei *N. volzi* bezeich-

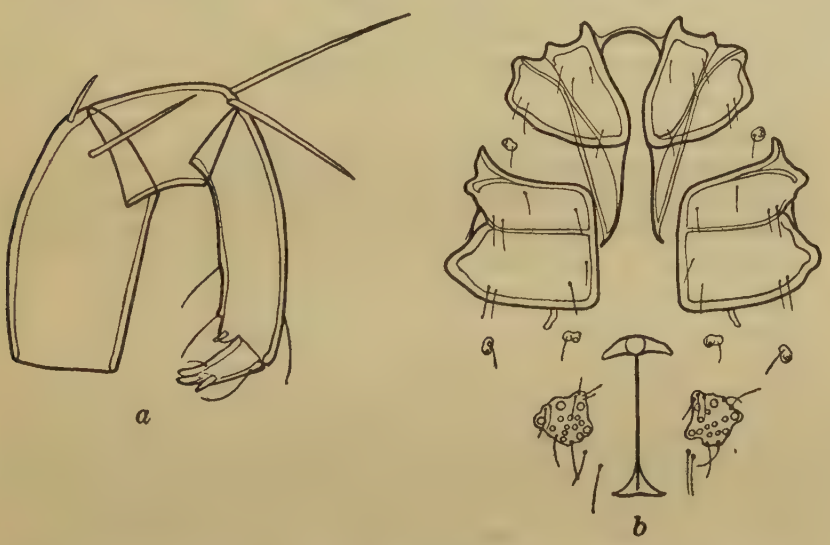

Fig. 2. *Neumania ambigua* Piersig; *a*, Weibchen, Palpus; *b*, Weibchen, Epimeralgebiet und Genitalorgan.

net, so ist das nur in bedingtem Malsse der Fall. So zählt man auf dem vierten Gliede der beiden Vorderbeine nur ein statt zwei Paare langer gerillter Degenborsten, auf dem fünften Gliede derselben nur zwei statt drei Paare. Ihr Endglied zeigt distal neben einer schwachen Biegung eine Verbreiterung vor dem Krallengrunde. Auf dem vierten und fünften Gliede des Hinterbeines sind alle Kurzborsten der Innenfläche gefiedert, die distale ist nur wenig länger als die übrigen; diese Dolchborsten nehmen regelmässig von der proximalen zur distalen an Länge zu. Der Schwimmhaarbesatz bei beiden Arten ähnlich, ebenso die Krallen. Das Endglied des Hinterbeines ist im Vergleich zu *N. volzi* relativ kürzer als der vorausgehende Artikel.

Piersig bildet in seiner Fig. 29 auf Taf. 15 das Genitalfeld eines noch nicht erwachsenen Weibchens ab, dessen Genitalöffnung am Hinterrande eine Einkerbung hervorruft und dessen Genitalplatten ganz an das Körperende zu liegen kommen. Bei ausgereiften Weibchen liegt aber das Genitalfeld ganz vom Körperrande ab. Genitalöffnung sehr lang (Fig. 2, *b*), bei gequetschten Tieren 0.255 Millimeter, bei einem Weibchen aus der Piersig'schen Sammlung, das noch die natürliche Wölbung der Bauchfläche zeigt, 0.185 Millimeter (nach Piersig bei einem noch jungen Exemplare 0.152 Millimeter). Genitalplatten klein, in nicht gewölbter Lage gleich weit vom vorderen wie vom hinteren Stützkörper entfernt. in natürlicher Lage dem hinteren scheinbar genähert. Ihre Form ist eine unregelmässig dreiseitige, ihr Rand infolge schwacher Chitinisierung unscharf, wellig. Nach Piersig sitzen auf jeder Platte 7 bis 11 Näpfe; die drei Exemplare aus seiner Sammlung tragen jederseits aber 13, 14, ja 18 Näpfe, das Philippinen-Exemplar auf der einen Platte 13, auf der andern 14. Auf den Plattenrändern, besonders in der Vorderecke der Platte zählt man vereinzelte kurze Haare, meistens auch zwei in der freien Haut hinter der Platte.

Excretionsporus am Körperhinterrande, etwas erhöht.

Fundort.—Philippinen, Luzon, ein Weibchen.

NEUMANIA FLAGELLATA sp. nov. Fig. 3.

Weibchen.—Die Körperlänge des im Umriss kurz-elliptischen Tieres beträgt 0.700 Millimeter bei einer Breite von 0.615 Millimeter. Stirnrand zwischen den antenniformen Borsten ganz schwach eingebogen. Diese sind kurz und fein, sitzen auf Nebenhöckern grosser Stirnzapfen und 225 μ voneinander entfernt. Alle Drüsenmündungen und die meisten Einzelhaare finden sich auf ähnlichen über die Haut hervorragenden Zapfen, wie auch der die Mitte des Hinterrandes einnehmende Excretionsporus. Haut ca. 15 μ dick, dicht mit feinen, sehr kurzen Chitinspitzchen besetzt. Die Augen liegen am seitlichen Vorderrande; sie sind schwarz pigmentiert. Beide Linsen eines Doppelauges sind elliptisch, die vordere mit 40 μ, die hintere mit 35 μ messendem grösstem Durchmesser.

Maxillarorgan und Mandibel je 105 μ lang, letztere mit stark hakenförmig gekrümmtem Klauenglied von 34 μ Länge. Palpus (Fig. 3, *a*) kurz und schwach; seine Glieder messen auf der Streckseite: Erstes, 18; zweites, 57; drittes, 28; viertes, 67; fünftes, 28 μ. Sie sind weiter charakterisiert durch sehr geringen Borstenbesatz, insbesondere durch die Abwesenheit langer, steifer Borsten am Distalende des dritten Gliedes und der

Tasthaarhöcker an der Beugeseite des vierten Gliedes. Tasthaare sehr fein und kurz, das äussere ganz distal, das innere nur wenig vom Gliedende abgerückt. Das vierte Glied trägt distal innen einen kurzen Chitinstift, aussen aber ein aussergewöhnlich langes und weiches Haar (über 0.3 Millimeter lang), das am Grunde ziemlich kräftig, gegen sein Ende aber sehr fein wird. Das Englied ist mit drei Klauen besetzt, von denen die dorsale etwas vom Gliedende abgerückt ist.

Epimeren vom Stirnrand abstehend, 0.360 Millimeter lang, 0.480 Millimeter breit. Ihre Oberfläche zeigt deutliche hexagonale Felderung, die einzelnen Plattengruppen sind breit umrandet. Epidermenidex = 3,65. Hinterrand der vierten Epimere transversal gerichtet, sein Fortsatz schwach entwickelt. Die äusseren Fortsätze der beiden hintern Platten nicht hakig (Fig. 3, *b*).

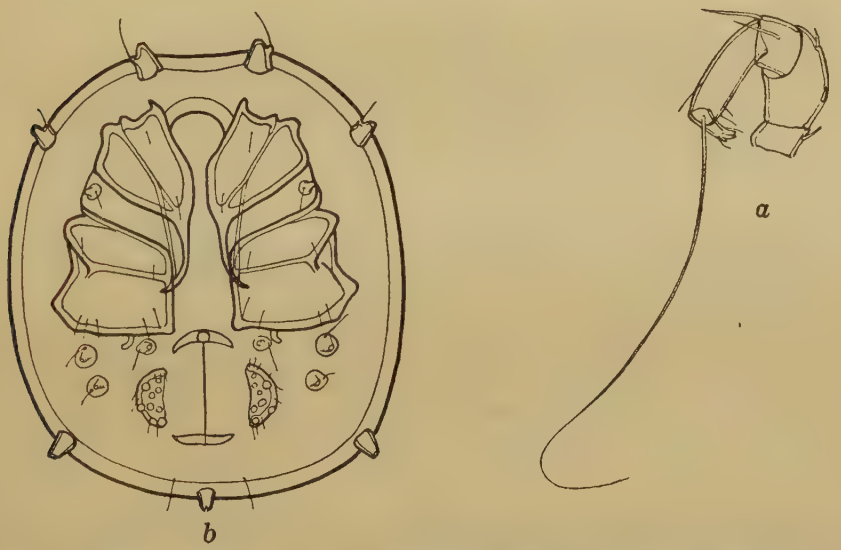

Fig. 3. *Neumania flagellata* sp. nov.; *a*, Männchen, Palpus; *b*, Männchen, Ventralansicht.

Beine im Vergleiche zu anderen *Neumania*-Spezies dünn, die Vorderbeine nur unbedeutend stärker als die nachfolgenden. Beinlängen: Erstes, 0.785; zweites, 0.810; drittes, 0.780; viertes, 0.900 Millimeter. Die gerillten Langborsten auf dem Vorderbeine wenig zahlreich, eine einzelne auf dem dritten, ein Paar auf dem vierten, zwei Paare auf dem fünften Gliede; auf dem zweiten Beine ein Paar auf dem vierten, zwei Paare auf dem fünften Gliede; sie sind jedoch kürzer als auf dem ersten Beine. Das fünfte Glied des dritten Beines trägt distal eine grobgefie-

derte Borste, auf die Gliedlänge verteilt 3–4 kurze Borsten, diese scheinbar ohne Fiederung. Einige wenige Schwimmhaare finden sich auf dem vierten und fünften Gliede des dritten Beines, auf dem dritten bis fünften Gliede des vierten. Innenfläche des Hinterbeines auf dem dritten Gliede mit 1, des vierten mit 3, des fünften mit 4 grobgefiederten kurzen Borsten. Kralle einfach, ohne Nebenzähne.

Das Genitalorgan ist nahe an das Epimeralgebiet gerückt. Die beiden Stützkörper gross, die Genitalspalte 180 μ lang. Jederseits derselben eine stark chitinisierte, am Aussenrande noch besonders verdickte Platte, die mit 8 Näpfen und wenigen feinen Randhaaren besetzt ist. Hinter der vierten Epimere liegen 3 Drüsenhöfe.

Fundort.—Philippinen, Luzon. Ein Weibchen, neun Eier von 0.170 Millimeter Durchmesser tragend.

ERKLÄRUNG DER FIGUREN

FIG. 1. *Limnesia bakeri* sp. nov.; *a*, Weibchen, Palpus; *b*, Männchen, Ventralansicht; *c*, Weibchen, Genitalorgan; *d*, Genitalplatte der Nymphe und die vier Näpfe des Teleiophanorganes.

2. *Neumania ambigua* Piersig; *a*, Weibchen, Palpus; *b*, Weibchen, Epimeralgebiet und Genitalorgan.

3. *Neumania flagellata* sp. nov.; *a*, Männchen, Palpus; *b*, Männchen, Ventralansicht.

NOTES ON PHLEBOTOMUS NICNIC BANKS

By C. Manalang
Of the Philippine Health Service, Manila

ONE PLATE

Banks [1] reported this species in 1919 as the first in the Philippines belonging to the genus *Phlebotomus* Rondani. Sinton [2] states the following about *P. nicnic* Banks:

> The male genitalia of this species resemble closely those of *P. minutus* Rond. and the possibility that it may be identical with *P. perturbans* de Meij. has been discussed under the latter species. There is also a possibility that it may be the same as some of the Indian species, such as *P. babu* Annandale, which have the same type of male genitalia. Until some more definite information is available as to the morphology of the buccal armature and other important diagnostic features, this species must be included in the list of Asiatic *Phlebotomus*. The species has only been recorded from the Philippine Islands.

In view of Sinton's doubts, the present study was undertaken with particular attention to the buccopharyngeal armatures [3] and the genitalia.[4] The results of the study seem to establish definitely that *P. nicnic* is a distinct species and not synonymous with *P. minutus, P. babu,* or *P. perturbans.*

Sand flies were obtained in the mornings from several anopheles mosquito-catching stations of the Novaliches water project, in May, 1929. A comparison of the specimens with the paratypes of *P. nicnic* Banks kindly furnished by Prof. L. B. Uichanco showed that I was dealing with the same species.

The method used for mounting the buccopharyngeal armatures is similar to the one used in the study of these structures in anopheles,[5] but the specimens were stained with carbol fuchsin. The spermathecæ were studied in fresh specimens, dissected in salt solution, and later preserved in 5 per cent formalin. The male hypopigium was observed in potash preparations.

[1] Philip. Journ. Sci. 14 (1919) 163–165.
[2] Ind. Journ. Med. Res. 16 (1928) 297-324.
[3] Adler, S., and O. Theodor, Bull. Ent. Res. 16 (1926) 399–405.
[4] Sinton, J. A., Ind. Journ. Med. Res. 15 (1927) 29–32.
[5] Manalang, C., Philip. Journ. Sci. 38 (1929) 131–136.

DESCRIPTION OF FEMALE

Medium sized, usually dark brownish gray, a few almost black (except the thorax); very hairy. Legs metallic silvery and wings and abdomen iridescent by transmitted light. Hairs on the clypeus, dorsum of the thorax, and first segment of the abdomen erect, on remaining segments recumbent; pleuræ without flat scales. Eyes black and oval.

Palps.—First segment short with two rows of stout hairs at the proximal end (Plate 1, fig. 1); second segment more than two times the first and shorter than the third or fourth; fifth segment slender, a little longer than the combined lengths of the third and fourth segments.[6] No modified spines. The segments bear scales and hairs. Total length of palpus, 0.72 millimeter.

Antennæ.—Second segment spherical, carrying a single verticil of stout long hairs (Plate 1, fig. 2); third segment two times the length of the fourth. All segments covered with hairs, sixteenth segment (Plate 1, fig. 3) with many short hairs. Short geniculate spines present; total length of the antenna, 1.17 to 1.24 millimeters.

Buccal armature.—Pigmented area cylindrical with slight expansion at its caudal half (Plate 1, fig. 8); two rows of minute, short, conical teeth, clearly seen only with oil-immersion lens. Pharyngeal bulb (Plate 1, fig. 9) flask-shaped, with three to four rows of slender, widely-separated teeth pointing backward among transverse ridges.

Thorax.—Integument colorless (hyaline in fresh specimen) laterally, dark brown to black dorsally; dorsum thickly covered with black vertical hairs; halteres black.

Abdomen.—Dorsum, except for first segment, covered with recumbent hairs. Integument almost black. Length, 0.90 to 1.2 millimeters; total body length, 1.88 to 2.231 millimeters.

Wings.—Hairy, lanceolate, and pointed (Plate 1, fig. 6). Third longitudinal vein dividing wing into two equal parts, the curvatures of the anterior and posterior borders the same. The upper or anterior branch of the second vein equals the distance between the two forks. Length, 1.600 to 1.650 millimeters; width, 0.500 millimeter.

[6] Banks's figure of the palp (Plate 1, fig. 1) and his description of it (p. 164) are apparently erroneous. He had the first and second segments considered as one segment, the first segment, and the fifth segment divided into two filiform segments which he called the 4th and 5th.

Hind legs.—Hind legs slightly longer than body, covered with hairs and scales arranged in bands, particularly on the tarsal segments (Plate 1, fig. 7). Ungues rectangular at base, body slender, slightly bent apically.

Genitalia.—Spermethecæ barrel-shaped and smooth, in one case they were very faintly striated (Plate 1, fig. 4). Spermathecal ducts join at acute angle (Plate 1, fig. 5).

DESCRIPTION OF MALE

Male slightly smaller than female; abdomen slender; antennæ slightly longer; palps shorter. Wings slightly shorter and considerably narrower; anterior branch of the second vein shorter than the distance between the two forks. Buccal and pharyngeal armatures similar to those of the female with the characteristic cylindrical pigmented area in the buccal cavity.

Genitalia.—Proximal segment of the superior clasper bears a thick brush of nondeciduous stout long hairs on the mesial surface; distal segment one-third the length of the proximal and armed with four stout spines (macrochætæ), three apical and one on a subapical tubercle, one or two spines with spatulate tips. Intermediate appendage simple finger-shaped, bearing hairs over its distal half. Intromittent organ paired but usually fused distally; Plate 1, fig. 11, shows them separated with tips of the genital filaments just protruding. Inferior claspers not armed. Plate 1, fig. 10, shows chitinous tube of "pompetta" and the two ejaculatory ducts.

DISCUSSION

Adler and Theodor's [7] figure of the spermatheca of *P. minutus* Rondani is similar to that of *P. nicnic* but the buccal and pharyngeal armatures are different, the pigmented area is oval with many long teeth in *P. minutus*, and cylindrical with minute short teeth in *P. nicnic*. Teeth in pharynx are numerous and point forward in *P. minutus*, but are few and point backward in *P. nicnic*. The buccal armature of *P. babu* Annandale, according to Sinton [5] has a mushroom-shaped pigmented area with depression in the top, and numerous teeth. The buccal armature of *P. perturbans* de Meijere, according to Patton and Hindle,[9] has a small, pale, somewhat semilunar pigmented area, and carries more, longer, and stouter teeth than *P. nicnic*.

[7] Loc. cit.
[5] Ind. Journ. Med. Res. 15 (1927) pl. 8, fig. 9; 16 (1928) 315.
[9] Proc. Royal Soc. London 102 Series B (1928) 542, fig. 9a.

I am unable to find figures or descriptions of the pharyngeal bulbs of *P. babu* and *P. perturbans* to compare with that of *P. nicnic*. However, the buccal armature of *P. nicnic* as described differs to such an extent from those which it simulates externally, that it is believed that *P. nicnic* should be considered a distinct species.

TABLE 1.—*Measurements of parts of the body.*

	Female 1.	Female 2.	Female 3.	Male 1.	Male 2.
	mm.	*mm.*	*mm.*	*mm.*	*mm.*
Body:					
Clypeus and head	0.330	0.356	0.350	----------	0.350
Thorax	0.500	0.550	0.500	0.450	0.430
Abdomen	0.900	1.200	1.200	1.200	1.100
Superior clasper, segment 1	0.150	0.125	0.150	0.225	0.237
Total length	1.880	2.231	2.200	----------	2.117
Labium	0.212	0.225	0.220	----------	0.175
Antenna:					
Segment III	0.158	0.162	0.162	0.212	0.206
Segment IV	0.075	0.081	0.075	0.100	0.100
Segment V	0.075	0.081	0.075	0.100	0.100
Segment VI	0.075	0.081	0.075	0.100	0.100
Segments XII to XVI	0.293	0.312	0.312	0.287	0.275
Total length (including segments I, II, and VII to XI)	1.176	1.247	1.174	1.381	1.399
Palp:					
Segment 1	0.037	0.037	0.037	0.041	0.037
Segment 2	0.093	0.100	0.100	0.087	0.081
Segment 3	0.137	0.125	0.125	0.125	0.137
Segment 4	0.143	0.150	0.137	0.150	0.137
Segment 5	0.318	0.312	0.300	0.275	0.260
Total length	0.728	0.724	0.699	0.678	0.652
Wing:					
Length	1.600	1.650	1.650	1.500	1.500
Breadth	----------	0.500	0.500	0.375	0.350
Hind leg:					
Femur	0.650	0.700	0.700	0.600	0.650
Tibia	0.800	0.850	0.900	0.720	0.760
Tarsus, segment 1	0.350	0.400	0.400	0.400	0.380
Tarsus, segments 2 to 5	0.550	0.600	0.580	0.550	0.525
Total length[a]	2.350	2.550	2.580	2.270	2.315
Superior clasper:					
Segment 1	----------	----------	----------	0.225	0.237
Segment 2	----------	----------	----------	0.075	0.087
Intromittent organ	----------	----------	----------	0.065	0.065

[a] Not including coxa and trochanter.

ILLUSTRATIONS

PLATE 1. PHLEBOTOMUS NICNIC BANKS.

FIG. 1. Female palpus (potash preparation mounted in Berlese's fluid).
2. Proximal portion of female antenna.
3. Distal portion of female antenna.
4. Spermatheca (dissection from a fresh specimen, preserved in 5 per cent formalin).
5. Junction of the two spermathecal ducts.
6. Denuded wing of female (potash preparation).
7. Last tarsal joints of hind leg of female (potash preparation mounted in Berlese's fluid).
8. Buccal armature of female (potash preparation stained with carbol fuchsin).
9. Pharyngeal armature of female (potash preparation stained with carbol fuchsin).
10. Chitinous ducts in the "pompetta" and the ejaculatory ducts (potash preparation).
11. Male genitalia (potash preparation); *m*, macrochætæ; *ds*, distal segment, superior clasper; *ps*, proximal segment, superior clasper; *b*, brush of hairs; *io*, intromittent organ; *ia*, intermediate appendage; *ed*, ejaculatory ducts; *ic*, inferior clasper.

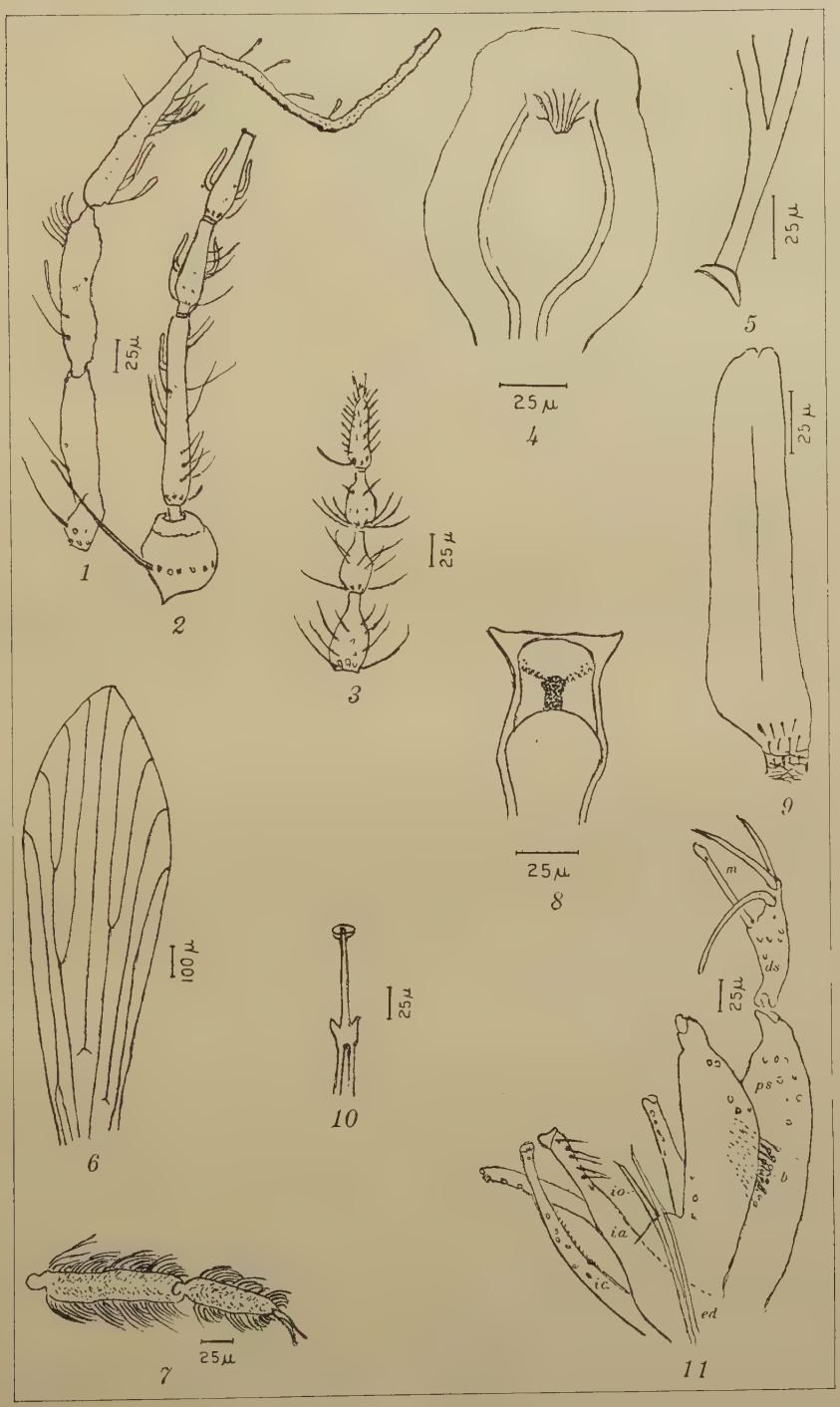

PLATE 1.

A NEW SPECIES OF THE GENUS PHLEBOTOMUS RONDANI

By C. MANALANG

Of the Philippine Health Service, Manila

ONE PLATE

Banks [1] published the first report in the Philippines of a species of this genus in 1919 and named it *Phlebotomus nicnic.*

The first specimen observed of this new sand fly was an erect-haired male in a collection from one of the mosquito stations of the Novaliches water project. It was among a number of *Culicoides* collected in April, 1929. In the collection made in May, after some rains, the *Culicoides* were replaced by sand flies, mostly of the new species, and a few recumbent-haired ones, *P. nicnic* Banks. After a week or so, *P. nicnic* outnumbered and finally entirely replaced the erect-haired flies in the collections. In July, the new species reappeared in the collections.

PHLEBOTOMUS PHILIPPINENSIS sp. nov. Plate 1.

DESCRIPTION OF FEMALE

Medium-sized, erect-haired, golden yellow with metallic luster on wings and legs when viewed by transmitted light. Eyes black, slightly oval to almost spherical. Clypeus and occiput with long vertical hairs.

Palps.—Characterized by short fourth segment (Plate 1, fig. 1) which is even shorter than the second; third segment almost as long as fifth; all segments covered with slender bent scales, but no modified spines are seen. Length, 0.53 millimeter.

Antennæ.—Second segment spherical with a single verticil of long hairs (Plate 1, fig. 2); third segment long, not quite reaching the tip of the labium, about two and a half times the fourth segment, rather densely covered, as the other segments, with long hairs; sensorium with shorter slender hairs (Plate 1, fig. 3). Long geniculate spines present. Length, 1.9 millimeters.

Buccal armature.—Without pigmented area but with fine, short, faintly-staining teeth arranged somewhat like a V (Plate

[1] Philip. Journ. Sci. 14 (1919) 163–165.

1, fig. 6). Pharyngeal bulb flask-shaped with many long stout teeth directed forward (Plate 1, fig. 9).

Thorax.—Integument pale yellow; no flat scales on the pleuræ; dorsum covered with long vertical hairs. Halteres long-stemmed, club-shaped, and covered with brown scales.

Abdomen.—Cylindrical, integument brownish yellow, dorsum with long vertical hairs arranged in tufts, some curved forward, others backward. Ventrum and sides covered with semirecumbent hairs. Length of abdomen, 1.2 millimeters. Total body length, 2.23 to 2.36 millimeters.

Wings.—Voluminous, covered with grayish brown hairs. Posterior border more curved than anterior (Plate 1, fig. 10). Upper or anterior branch of the second longitudinal vein almost twice as long as the distance between the two forks. Wing, 1.80 by 0.7 millimeter.

Hind legs.—Long, covered with scales and hairs distinctly arranged in bands particularly on the tarsal segments (Plate 1, fig. 8). Ungues slender and straight. Length of hind legs, 3.43 millimeters; about one and one-fourth times the length of the body.

Genitalia.—Spermathecæ carrot-shaped, with eleven crenulations, small head, and short neck (Plate 1, fig. 4). The spermathecal ducts join at acute angle into a common highly chitinized yellowish duct (Plate 1, fig. 5).

DESCRIPTION OF MALE

Abdomen slenderer than in female, and terminalia bent dorsally, size and color about the same; the abdominal integument of female somewhat darker. Buccal armature like that of female; pharyngeal bulb without the long, stout teeth seen in the female but with transverse chitinous ridges only.

Genitalia.—Distal segment of the superior clasper slightly less than one-half the length of the proximal segment, and armed with five stout pointed spines (marcrochætæ) placed two apically, two subapically, and one just below the mid-portion (Plate 1, fig. 7). Intermediate appendage with three lobes and a long stout spine, the spine longer than the intromittent organ. The inferior lobe a broad pigmented prominence, bearing four hairs (Plate 1, fig. 11). Length of intromittent organ, 0.08 to 0.087 millimeter, the genital filaments hardly protruding. Inferior claspers not armed. Chitinous tube of the "pompetta" and the two ejaculatory ducts are shown on Plate 1, fig. 12.

TABLE 1.—*Measurements of parts of the body of Phlebotonus philippinensis sp. nov.*

	Male 1.	Male 2.	Female 1.	Female 2.
Body:	*mm.*	*mm.*	*mm.*	*mm.*
Clypeus and head	0.330	0.400	0.430	0.400
Thorax	0.450	0.500	0.575	0.500
Abdomen	1.400	1.100	1.200	1.200
Superior clasper, segment 1	0.225	0.225	0.160	0.130
Total length	2.405	2.225	2.365	2.230
Labium	----------	0.250	0.300	----------
Antenna:				
Segment III	0.337	0.325	0.300	0.275
Segment IV	0.131	0.137	0.125	0.125
Segment V	0.131	0.137	0.125	0.125
Segment VI	0.131	0.137	0.125	0.125
Segments XII to XVI	0.450	0.450	0.450	0.470
Total length (including segments I, II, and VII to XI)	1.985	2.021	1.900	1.895
Palp:				
Segment 1	0.050	0.037	0.037	0.037
Segment 2	0.087	0.087	0.100	0.110
Segment 3	0.125	0.125	0.158	0.150
Segment 4	0.062	0.062	0.075	0.075
Segment 5	0.137	0.150	0.160	0.162
Total length	0.461	0.461	0.530	0.534
Wing:				
Length	1.750	1.700	1.800	1.800
Breadth	0.530	0.570	0.680	0.700
Hind leg:				
Femur	0.660	0.750	0.750	0.675
Tibia	1.220	1.250	1.250	1.250
Tarsus, segment 1	0.700	0.730	0.730	0.750
Tarsus, segments 2 to 5	0.700	0.750	0.700	0.750
Total length [a]	3.280	3.480	3.430	3.425
Superior clasper:				
Segment 1	0.225	0.225	----------	----------
Segment 2	0.100	0.100	----------	----------
Intromittent organ	0.080	0.087	----------	----------

[a] Not including coxa and trochanter.

DIFFERENTIAL DIAGNOSIS

This species is easily differentiated from *P. nicnic* by the erect hairs on the dorsum of the abdomen (*P. nicnic*, recumbent), the broader wings, and five spines (macrochætæ) on the distal segment of the superior clasper of the male (*P. nicnic* has four). The spermathecæ of this species are similar to those of *P. argentipes* Annandale, but with marked differences in the pharyngeal

armatures.[2] In *P. argentipes* the teeth in the pharynx are few and point backwards.

Phlebotomus stantoni Newstead differs from the new species because, according to Sinton,[3] the pharyngeal armature of this species is similar to that of *P. argentipes.*

[2] Sinton, J. A., and J. P. Barraud, Ind. Journ. Med. Res. 16 (1928) pl. 29, fig. 5; pl. 30, fig. 9.

[3] Ind. Journ. Med. Res. 16 (1928) 297–323.

ILLUSTRATIONS

PLATE 1. PHLEBOTOMUS PHILIPPINENSIS SP. NOV.

FIG. 1. Female palpus (potash preparation mounted in Berlese's fluid).
2. Proximal portion of female antenna.
3. Distal portion of female antenna.
4. Spermatheca (preserved in 5 per cent formalin).
5. Junction of the two spermathecal ducts.
6. Buccal armature, female (potash preparation, stained with carbol fuchsin).
7. Male genitalia (potash preparation); *ed*, ejaculatory ducts; *sc*, superior clasper; *ps*, proximal segment, superior clasper; *ds*, distal segment, superior clasper; *m*, macrochætæ; *ic*, inferior clasper; *ia*, intermediate appendage; *s*, spine of the intermediate appendage.
8. Last tarsal segments, hind leg, female (potash preparation, mounted in Berlese's fluid).
9. Pharyngeal armature (potash preparation, stained).
10. Denuded wing of female (potash preparation).
11. Intermediate lobe of male genitalia (potash preparation); *1*, main lobe; *2*, middle lobe; *3*, inferior lobe; *s*, spine; *io*, intromittent organ.
12. Chitinous ducts in the "pompetta" and ejaculatory ducts.

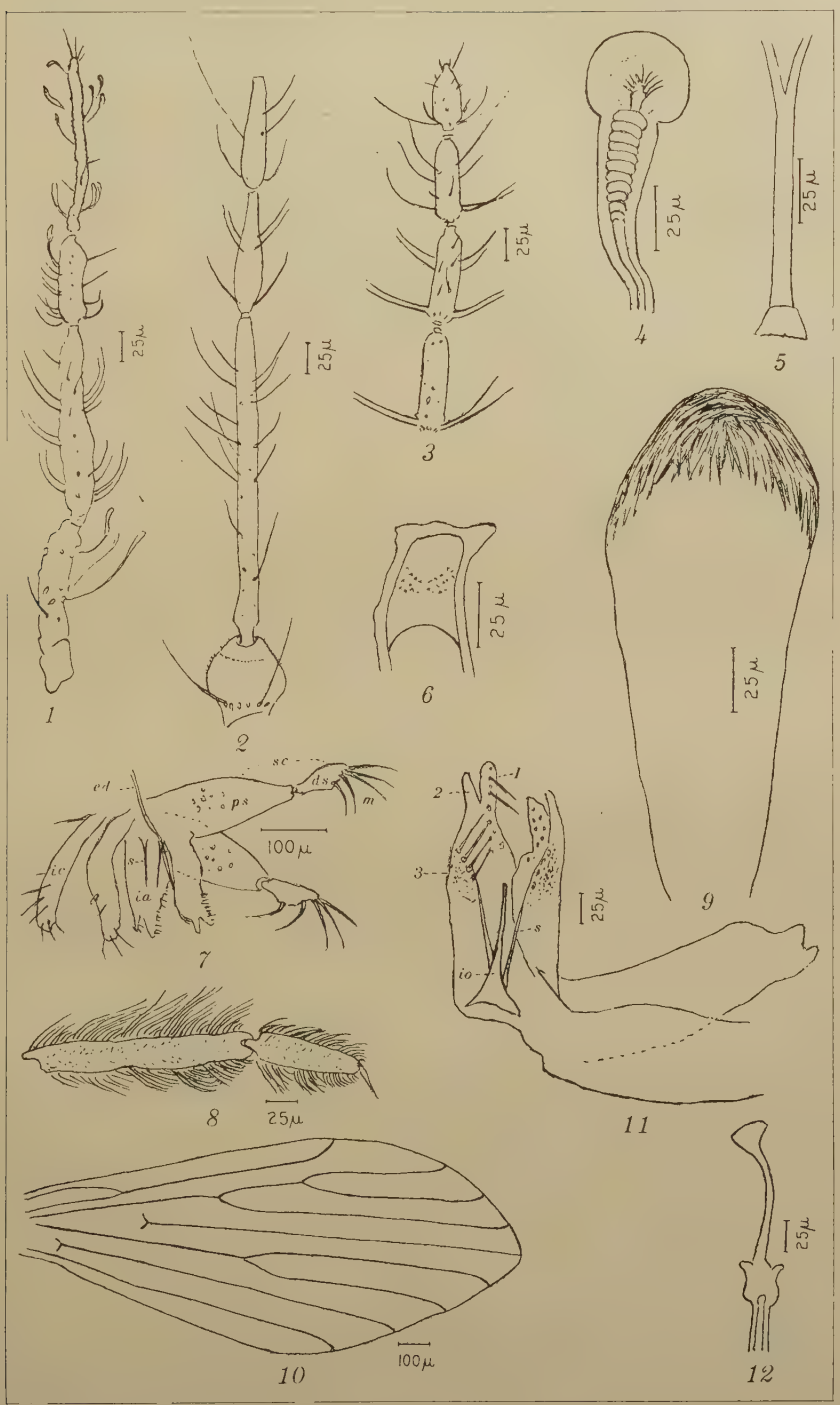

PLATE 1.

A TAXONOMIC STUDY OF PSEUDOMONAS SUIS ISOLATED FROM CROUPOUS PNEUMONIA IN SWINE [1]

By GLADYS M. WOODS

Of the Division of Veterinary Science, University of California, Berkeley

SIX PLATES AND SIX TEXT FIGURES

PSEUDOMONAS SUIS sp. nov.

The microörganism concerned in the taxonomic study described in this paper was isolated in the Philippine Islands by Dr. William H. Boynton, who kindly furnished the following history. In June, 1916, an outbreak of suspected swine plague occurred in hogs owned by the Philippine Vegetable Oil Company, just outside of Manila. The loss was heavy; it was estimated that of about one hundred fifty hogs, the mortality reached 50 per cent. The affected animals showed marked emaciation, suggesting a chronic type of disease. The predominating lesions, upon autopsy of some of these hogs, were those of croupous pneumonia and pleurisy. Cultures made from lung tissue revealed a rodlike organism which proved to be motile, and, therefore, was not the swine-plague organism. A month later, three dead animals from the same herd were autopsied, and lung lesions were found similar to those of the previous cases. The organism was, to all appearances, the same. Again, in 1918, a case occurred, in Alabang, Rizal Province, Philippine Islands. This case was a sow that had been imported from the United States. The lesions in this case, and the organism, were similar to those of the first outbreak. From another case, which was reported March 10, 1920, and suffered an infection of *Balantidium coli* along with the pneumonia, the same organism was isolated as in the previous cases. The fact that the organism was isolated repeatedly suggested that it was the etiologic agent responsible for this pneumonic disease and the problem of determining its taxonomic relationship presented itself.

[1] Submitted in partial requirement for the master's degree under the direction of Dr. Karl F. Meyer, Department of Bacteriology, University of California, Berkeley.

The strain used in this work is a passage strain of one which was isolated from the first outbreak, and transferred from time to time on artificial media. A suspension of culture was first injected intravenously, December 9, 1918, into a hog which died within twenty-four hours, too soon for characteristic lesions to develop. However, the organism was isolated from the heart, lungs, liver, spleen, kidney, and lymphatics, but not from the urinary bladder. A culture obtained from this animal was inoculated into two guinea pigs August 27, 1923; the culture at that time was 4 years 8 months 17 days old. These two guinea pigs died August 30, 1923, and the organism was recovered from the heart, lungs, liver, spleen, and urinary bladder. The hearts, lungs, livers, and spleens of these animals were fed to two hogs, 201–8 and 1, which died on the seventeenth and twenty-third days, respectively. The animals showed marked lung lesions and necrotic foci in the liver and spleen. The organism was recovered from the hearts, lungs, livers, spleens, and kidneys of these hogs. Strain 201–8 was given to me for study.

MORPHOLOGICAL STUDY OF THE ORGANISM

This organism is a small rod with either rounded or pointed ends; at times it is slightly curved and in old cultures it frequently appears almost coccoid. In length it varies from 1.2 to 3.6 microns; in thickness, from 0.5 to 1.5 microns. There appears to be no chain formation. It stains readily with the ordinary aniline dyes, is discolored by the Gram method, and frequently shows bipolar staining. This latter characteristic was at first thought to be noticeable only in cultures several days old or in those freshly isolated from animals, but the same tendency has also been observed in ordinary twenty-four-hour transfers. The organism is not acid-fast and shows neither spores nor capsules. In twenty-four-hour cultures it is actively motile. There was some question as to whether the flagella were lophotrichous, but of those examined, the greater number were found to be monotrichous; the majority of those which showed flagella at both poles were observed to be dividing. The number of flagella varies from one to five (text fig. 1).

Cultural characteristics.—In general, the methods followed were those compiled in the Manual of Methods for Pure Culture Study of Bacteria.(1)

Gelatin plates: White, pinhead colonies appeared on the plates in forty-eight hours at room temperature. Microscopically, these were granular in substance, recticulate-moruloid in

type, with a lobar-lobulate edge. In seventy-two hours, depressions were observed around the colonies, indicating that liquefaction was taking place. Under the microscope, these colonies were shown to be disintegrating. They were less lobed in appearance and contained larger granules.

Gelatin stab: After twenty-four hours, there was no growth along the line of stab. In seventy-two hours, growth developed at the surface of the medium with crateriform liquefaction. Within ten days, the liquefaction deepened to the stratiform type.

Agar plates: The colonies appeared grayish white, round, smooth, translucent, rather glistening, and slightly raised, after

FIG. 1. The swine-pneumonia organism, showing flagella. (After Pitfield.)

twenty-four hours. In seventy-two hours, they became a dirty cream in color, opaque, and wrinkled, with an undulate edge. Microscopically, the colonies consisted of a browinsh yellow, nongranular substance with a small, central, brown, elliptical mass, and a very slightly undulate edge. After seventy-two hours, they appeared more granular, showed radial folds, and a more markedly undulate border. On agar which contained 1 per cent glucose or 2 per cent glycerin and 1 per cent glucose the wrinkling or folding characteristic appeared a little earlier, in forty-eight hours. On agar containing defibrinated horse blood, the colonies had a similar appearance, and hæmolysis of the beta type occurred in forty-eight hours. That this hæm-

olysis is due to some other agent than a true hæmolysin is suggested by the fact that hæmolysis did not occur in cultures grown in yeast-infusion media containing blood. To substantiate the hypothesis that agents other than a true hæmolysin may be responsible for hæmolysis, there is the report of Orcutt and Howe(2) that the hæmolytic action of a staphylococcus was due to a fat-splitting enzyme.

Agar slant: Abundant, whitish, glistening, translucent growth appeared in twenty-four hours. It did not assume a wrinkled appearance unless glucose or glycerin and glucose were present in the medium. Then the growth was cream-colored and dry, with folds around the margin; later the entire streak appeared wrinkled. This is more characteristic of cultures freshly isolated from animals than of transfer cultures. On cultures a month old or older the growth frequently assumed a slightly brick-red coloration; and a not unpleasant yeastlike odor is characteristic of cultures that have stood for such a period of time.

Agar stab: A smooth, rather dirty, glistening, slightly raised growth appeared on the surface of the medium. Along the line of stab, growth was of the echinate type.

Bouillon: After twenty-four hours, uniform turbidity was observed; there was a thin white surface film and a slight amount of sediment. In bouillon containing glucose or glycerin and glucose a heavy wrinkled pellicle appeared. Upon shaking, the pellicle broke up and sank to the bottom of the tube; after twenty-four hours, a pellicle reappeared at the surface.

Peptone solution: In 1 per cent peptone water there was turbidity and a surface film in twenty-four hours. No stalactite formation could be observed after six days in cultures grown in peptone solution covered with a layer of oil.

Potato culture: Fairly abundant, smooth, rather moist, creamish to yellow-brown growth appeared in forty-eight hours. On potato medium to which 5 per cent glycerin had been added, growth was yellowish brown and dry.

Physiological characters.—Endo medium: Colonies were gray, rounded, and translucent in twenty-four hours. They appeared reddish in forty-eight hours, due possibly to alkali rather than acid formation, since in lactose broth medium the hydrogen-ion concentration changes from P_H 7.2 to P_H 7.8 in forty-eight hours.

Brilliant green agar: In twenty-four hours, colonies appeared. They were yellow and the surrounding medium was partially

changed to yellow, indicating reduction of the dye. Colonies became a dirty yellow in seventy-two hours. The dye apparently had no inhibitive effect upon the growth of the organism.

Litmus lactose agar: Gray colonies appeared in twenty-four hours. No change in color was produced upon further incubation.

Conradi-Drigalsky agar: Grayish blue colonies were observed in twenty-four hours.

Neutral red agar: Colonies in twenty-four hours were pink. In forty-eight hours the colonies became yellowish brown, indicating reduction of the dye.

Litmus milk: The medium was decolorized in forty-eight hours, showing that reduction of the litmus had taken place. Within seventy-two hours, the milk was coagulated. Peptonization occurred and the medium tended to become alkaline in fourteen days. Plain milk with a P_H of 6.0 at the time of inoculation changed to P_H 8.0 within fourteen days.

Indol was not produced in Dunham's peptone solution.

Production of hydrogen sulphide could not be observed in lead acetate medium after seven days.

Nitrites were produced, but no gas, in peptone solution containing nitrate.

Enzyme action: No diastatic action was produced on starch agar plates.

Gelatinase was demonstrated in gelatin culture filtrate. It produced liquefaction of gelatin within thirteen to twenty days.

Fermentation of carbohydrates: The medium used was a 1 per cent solution of Witte's peptone to which 1 per cent of each carbohydrate was added. This afforded more-delicate results than a broth medium. The reactions are shown in Table 1.

It will be seen from Table 1 that only two of the hexoses, dextrose and galactose, were definitely fermented. There was some slight degree of acidity in arabinose and a slighter degree in mannose, maltose, mannite, dulcite, and glycerin; but this changed back towards the alkaline side.

Gas production has never been observed in any of the carbohydrates.

Oxygen requirements: Agar shake cultures showed growth only on the surface of the medium. Bouillon cultures, grown anaërobically by Wright's method of using pyrogallic acid and potassium hydroxide, appeared to grow rather abundantly, but slant cultures grown by this same method were scanty. This would indicate that the organism is facultatively anaërobic.

TABLE 1.—*Fermentation of carbohydrates.*

[The minus sign indicates a higher acidity than P_H 4.7. No buffer with a P_H value lower than 4.7 was available.]

Carbohydrate.	P_H value.								
	P_H before inoculation.	Strain 201–8.				Strain 3912 (guinea-pig passage strain of 201–8).			
		24 hours.	48 hours.	5 days.	10 days.	24 hours.	48 hours.	5 days.	10 days.
Dextrose	7.2	5.8	5.0	4.7	—4.7	6.2	5.3	—4.7	—4.7
Levulose	7.4	7.2	7.0	7.2	7.6	7.2	7.0	7.2	7.6
Galactose	7.4	6.0	5.0	—4.7	—4.7	6.6	5.8	—4.7	—4.7
Mannose	7.4	7.0	6.6	6.6	7.4	7.0	6.6	6.6	7.2
Saccharose	7.2	7.2	7.2	7.6	8.0	7.2	7.4	7.8	8.2
Maltose	7.2	7.0	6.6	6.8	7.4	7.0	6.6	6.6	6.8
Lactose	7.2	7.2	7.0	7.0	7.6	7.2	7.2	7.4	7.6
Arabinose	7.2	7.0	6.6	6.2	7.0	6.8	6.6	6.0	6.6
Xylose	7.2	7.2	7.2	7.2	7.6	7.0	7.0	7.2	7.6
Dextrin	7.2	7.2	7.2	7.4	8.2	7.2	7.2	7.6	8.2
Mannite	7.2	7.0	6.8	6.8	7.4	7.0	6.8	6.8	7.4
Dulcite	7.4	7.2	6.8	6.8	7.6	6.8	6.6	6.6	7.4
Glycerin	7.4	7.2	6.8	7.0	7.4	7.0	6.8	6.8	7.4

Temperature requirements: In this experiment 0.01 cubic centimeter of broth culture containing approximately 10,000 organisms was inoculated into a series of tubes containing 10 cubic centimeters of beef extract broth, P_H 7.4. These tubes were incubated at four temperatures, 0, 21, 39, and 42 to 45° C., for twenty-four hours. At the end of this time, 0.01 cubic centimeter was diluted in 10 cubic centimeters of physiological saline and 0.01 cubic centimeter of this suspension was plated. Colonies on the plates were counted following a forty-eight hour incubation. The results obtained are shown in Table 2.

TABLE 2.—*Optimum temperature for growth.*

°C.	Colonies per cubic centimeter.
0	50,000
21	50,000
39	111,500,000
42–45	30,500,000

Table 2 suggests that the optimum temperature is near 37° C., or body temperature.

Hydrogen-ion requirements: The general method used in the determination of the optimum hydrogen-ion concentration for growth was the one outlined by Schoenholz and Meyer.(3) Salt-free bouillon was used to which varying amounts of acid, alkali,

and buffer solution were added to give reactions ranging from P_H —5.0 to P_H 8.6. Into each tube of this series containing 5 cubic centimeters of a definite P_H value, 0.1 cubic centimeter of a 1 : 10,000 dilution of twenty-four-hour culture was inoculated. After twenty-four hours incubation, these cultures were plated; the inoculum was 0.1 cubic centimeter of a 1 : 100,000 dilution. The plates were counted following a twenty-four hour incubation period. The results are given in Table 3.

TABLE 3.—*Optimum hydrogen-ion concentration for growth.*

[—5.0 means beyond P_H 5.0 in the acid range. No indicator solution with a P_H value of less than 5.0 was available.]

P_H of medium.		Number of colonies in 24 hours in 1.0 cubic centimeter.	P_H of medium.		Number of colonies in 24 hours in 1.0 cubic centimeter.
Before inoculation.	24 hours after inoculation.		Before inoculation.	24 hours after inoculation.	
—5.0	5.1	21,000,000,000	6.9	6.9	7,100,000,000
5.1	5.3	12,760,000,000	7.1	7.1	6,130,000,000
5.7	5.9	53,990,000,000	7.3	7.3	6,100,000,000
5.9	6.0	40,830,000,000	7.5	7.5	4,200,000,000
6.1	6.3	59,500,000,000	7.7	7.7	2,860,000,000
6.3	6.4	8,160,000,000	7.9	7.9	820,000,000
6.5	6.5	16,200,000,000	8.2	8.2	10,000,000
6.7	6.7	11,060,000,000	8.6	8.6	None

It will be noted in Table 3 that the numbers of colonies counted for certain hydrogen-ion concentrations do not show a proprotional increase or decrease, but repeated tests showed the same discrepancies which are probably due to difficulties in breaking up clumps of bacteria held together by their flagella. The logarithms of the numbers of colonies are plotted as ordinates (text fig. 2) against the hydrogen-ion concentrations as abscissæ, to obtain the growth curve, which demonstrates that the organism prefers the acid range and that the optimum point is about P_H 6.1.

PATHOGENICITY

No experiments on hogs were carried out in the United States. Aside from the apparently natural infection in swine, Boynton, as was mentioned in the history of the present strain, was

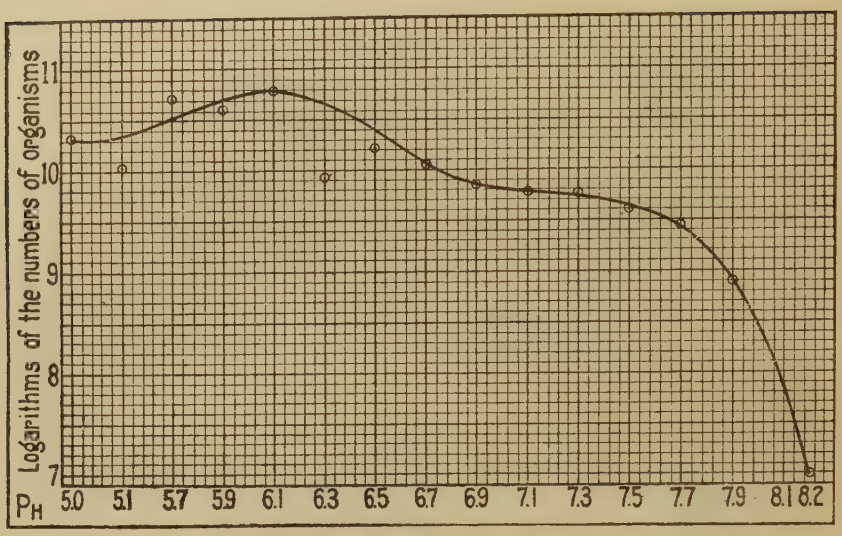

FIG. 2. Growth curve of the organism.

able to transmit the disease to a hog by intravenous inoculation of a suspension of culture. Death of the animal occurred within twenty-four hours, too rapidly for lesions to develop. The organism was isolated from the liver, spleen, heart, lungs, and lymphatics. Two hogs, 201–8 and 1, were fed the hearts, lungs, livers, and spleens of two infected guinea pigs and they died on the sixteenth and twenty-second day, respectively, following the feeding. The temperature curves of these animals were given to me to examine and are reproduced in text fig. 3.

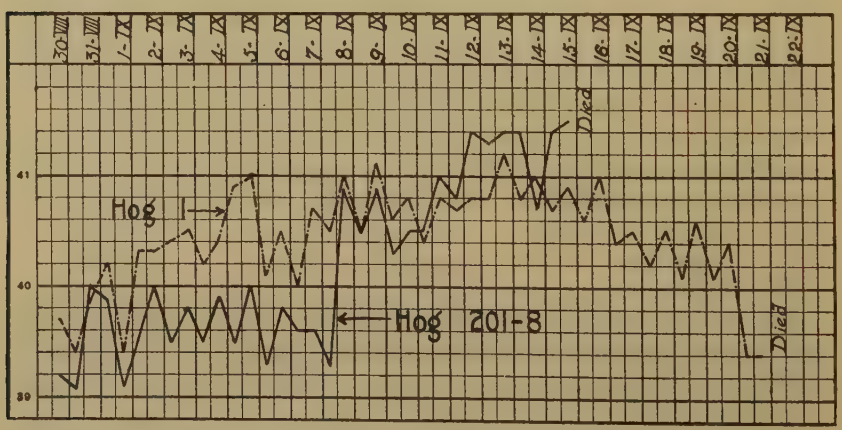

FIG. 3. Temperature curve of two hogs which were fed livers, hearts, spleens, and lungs of two guinea pigs infected with the swine-pneumonia organism, 30–VIII–23.

Among the laboratory animals this organism is pathogenic to rabbits, guinea pigs, and mice. It is strongly virulent for these species, especially for rabbits, and the virulence does not appear to change appreciably. Rats may be susceptible to a slight degree. Pigeons are sometimes affected. The single chicken tested failed to show infection. The pathogenic effects upon these species will be discussed in the order they are named.

In rabbits, infection was established by the three routes tested, as shown in Table 4.

The smallest dose tested was an attempt to immunize by means of an unattenuated vaccine. This dose was 0.1 cubic centimeter of a 1: 100,000 dilution of broth culture and it contained approximately 33,600 organisms per cubic centimeter. It was administered subcutaneously and killed the rabbit in six days.

TABLE 4.—*Pathogenicity experiments on rabbits.*

Route of infection.	Dosage.	Result.	Post-mortem findings.	Bacteriological findings.
Intravenous; animal 1.	0.2 cubic centimeter of saline suspension of agar culture: 3–IX–24.	Died in twenty hours.	Engorgement of spleen.	Organism recovered from heart blood, lung, liver, spleen, kidney, portal lymph gland, and bone marrow.
Subcutaneous; animal 2.	0.4 cubic centimeter of same suspension: 3–IX–24.	Died in forty-eight hours.	Necrotic foci in liver and spleen. Inflammation at point of injection.	Organism recovered from heart blood, lung, liver, spleen, kidney, portal and submaxillary lymph glands, and bone marrow.
Subcutaneous; animals 3 and 4.	0.5 cubic centimeter of 1–10 dilution of saline suspension of agar culture; Gates reading 2.1: 15–IV–25.	...do.....	Necrotic foci in liver and spleen. Slight hæmorrhage and abscess formation at point of injection.	Organism recovered from heart blood, liver, and spleen.
Subcutaneous; animal 5.	0.1 cubic centimeter of 1:100 dilution of broth culture containing approximately 33,600,000 organisms per cubic centimeter: 5–XII–25.	Died in three days.	Necrotic foci in spleen, liver, and lungs.	Organism recovered from lung and spleen.

TABLE 4.—*Pathogenicity experiments on rabbits*—Continued.

Route of infection.	Dosage.	Result.	Post-mortem findings.	Bacteriological findings.
Oral; animal 6.	.01 cubic centimeter of 1:10 dilution of broth culture containing approximately 336,000,000 organisms per cubic centimeter: 5–XII–26.	Died two days after last feeding.	Necrotic foci in lungs and spleen. Few foci in liver.	Organism recovered from heart blood, lung, liver, and spleen.
	0.5 cubic centimeter of 1:10 dilution of broth culture containing approximately 336,000,000 organisms per cubic centimeter: 12–XII–26.			
	1 cubic centimeter of 1:10 dilution of broth culture containing approximately 336,000,000 organisms per cubic centimeter: 19–XII–26.			
	2 cubic centimeters of 1:10 dilution of broth culture containing approximately 336,000,000 organisms per cubic centimeter: 24–XII–26.			

The symptoms produced are rise in temperature and loss of weight. The increase in temperature occurs usually on the following day and remains continuous until the animal succumbs. (The temperature kept on a fed rabbit failed to rise. Since it remained normal and the animal did not continue to lose weight, these records were discontinued. On the thirty-third day, however, the animal was found moribund.) A typical case is shown in text fig. 4; the decrease in weight is also indicated. Rabbits which survive several days lose their appetite and become greatly emaciated. Toward the last, labored breathing is sometimes noticeable.

Post-mortem examination, as indicated in Table 4, revealed no change except slight engorgement of the spleen in an animal which succumbed to an intravenous dose of culture within twenty-four hours. In cases which survive forty-eight hours, the liver and spleen are studded with necrotic foci. Animals that live

longer than forty-eight hours usually show necrotic foci in the lungs. The longer they survive the more pronounced are the lesions, especially in the lungs and spleen. The foci protrude slightly on the surface of these organs and, on the cut surface, appear as spherical masses about 1 millimeter in diameter. In one case, definite enlargement of the spleen was observed. Necrotic foci were observed only once in the kidney; this was in a chronic case. The area around the point of inoculation frequently appears hæmorrhagic and shows abscess formation.

Bacteriological examination shows the organism to be almost invariably present in the liver and spleen. It is generally found also in the lungs and heart blood, to a less degree in the kidney, and it has been isolated once from the testicle, uterus, urinary bladder, and submaxillary lymph gland; and twice from the portal lymph gland and bone-marrow.

Table 5 shows that guinea pigs were infected by the subcutaneous and oral methods, and in three cases by smearing the nostrils with a swab dipped in a suspension of twenty-four hour agar culture.

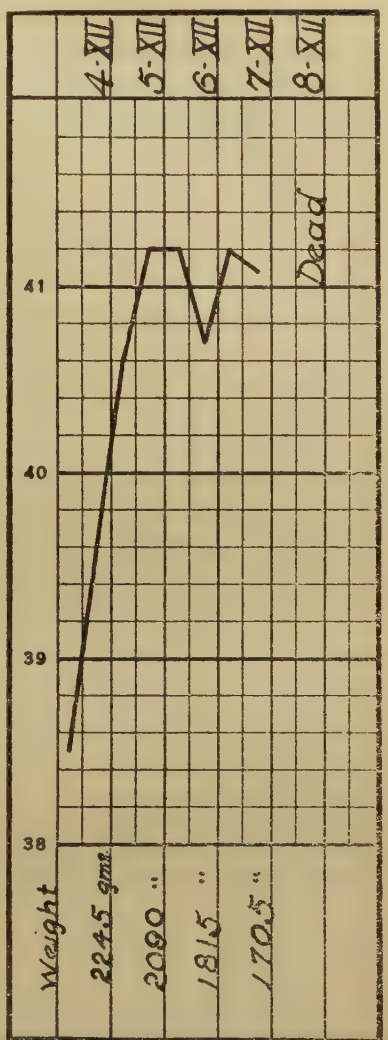

FIG. 4. Temperature curve of a rabbit which received subcutaneously 0.2 cubic centimeter of a 1:10 dilution of 16-hour broth culture containing approximately 85,000,000 organisms, 4–XII–26.

The effect of the organism upon these animals is much the same as upon rabbits. Rise of temperature and loss of weight are the chief symptoms noted, as text fig. 5 indicates. The effect of small doses is a more chronic type of infection, and this was best illustrated in the guinea pig. Inappetence and emaciation occur. In some cases, inflammatory changes in the eye have been observed, in which a cream-colored, purulent

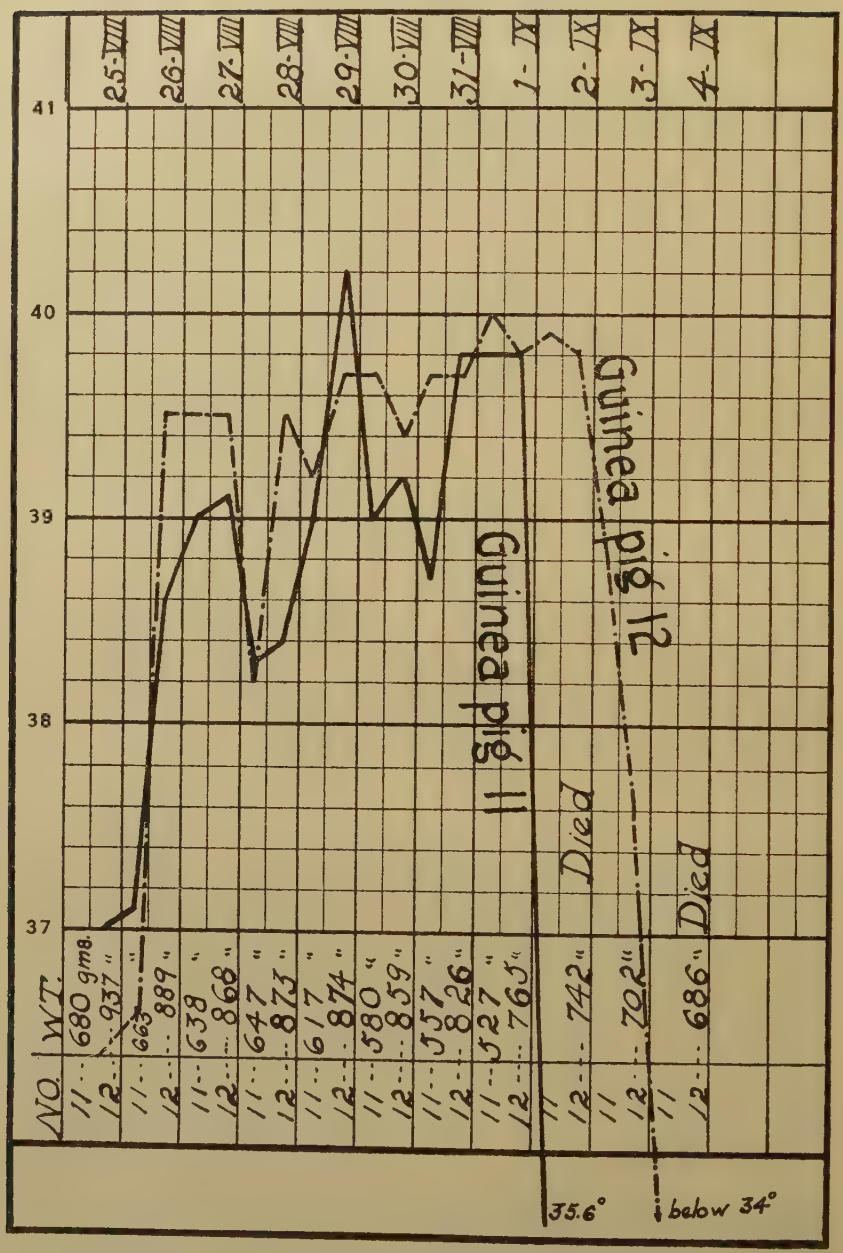

FIG. 5. Temperature curve of two guinea pigs inoculated by smearing their nostrils with a swab dipped in a broth suspension of a 24-hour agar culture, 25-VIII-26.

discharge forms, exudates, and becomes encrusted. The eye becomes sensitive to light and finally closes. Abscesses occur at and near the point of injection, later becoming open ulcers, and metastases may develop (Plate 1); and occasionally the hind legs drag. In the last stages, respiration becomes very labored and distinctly audible.

Post-mortem examination shows the same type of necrotic foci as found in the rabbit. The foci occur in the liver and spleen and then in the lungs (Plates 2, 3, and 4). In chronic cases, the foci are more numerous and more pronounced, as in the rabbit. The spleen usually becomes enlarged to about twice its normal size; the lungs show hepatization and foci 1 to 1.5 millimeters in diameter. These characteristic necrotic spots were observed only once by me in the heart and kidney, although Boynton reports finding them previous to this work. Abscesses occurring at the point of inoculation and in the inguinal glands may measure 1 to 2 centimeters in diameter. In one case, an abscess about 1 centimeter in diameter was found in the pectoral gland above the point of injection. Orchitis has occurred in a few instances.

TABLE 5.—*Pathogenicity experiments on guinea pigs.*

Route of infection.	Dosage.	Result.	Post-mortem findings.	Bacteriological findings.
Subcutaneous; animals 3911 and 3912.	0.4 cubic centimeter of saline suspension of agar culture from 201–8: 21–VIII–24.	Died in seventy-two hours.	Inflammation at point of injection. Necrotic foci in lungs, liver, and spleen. Foci in heart and kidney of 3911.	Organism recovered from lung, liver, spleen, kidney, and urinary bladder. Also from heart blood of 3911.
Subcutaneous; animal 3.	0.4 cubic centimeter of saline suspension of culture from 3911: 3–IX–24.	Killed moribund in forty-eight hours.	Necrotic foci in liver and spleen. Inflammation at point of injection.	Organism recovered from heart blood, lung, liver, spleen, kidney, urinary bladder, and bone-marrow.
Subcutaneous; animal 1LD	0.5 cubic centimeter of a 1:100,000 dilution of broth culture containing 17,600 organisms per cubic centimeter: 1I–1–27.	Died in eight days.	Spleen enlarged to twice normal size. Numerous necrotic foci in spleen and liver. Orchitis present. Abscesses at point of injection (left) and in right inguinal gland. Pronounced lesions in lungs.	Organism recovered from heart blood, lung, liver, spleen, and testicle.

TABLE 5.—*Pathogenicity experiments on guinea pigs*—Continued.

Route of infection.	Dosage.	Result.	Post-mortem findings.	Bacteriological findings.
Subcutaneous; animal 2LD	0.1 cubic centimeter of a 1:10,000 dilution of same culture (176,000 organisms per cubic centimeter).	Died in eight days.	Pronounced lesions in lungs, liver, and spleen. Spleen enlarged to twice normal size. Abscesses at point of injection (left) and right inguinal gland.	Organism recovered from lung and spleen.
Subcutaneous; animal 3LD	0.5 cubic centimeter of 1:10,000 dilution.	Died in eighteen days.	Gray hepatization and few necrotic foci in lungs. Necrotic patches in liver and pronounced foci in spleen. Spleen enlarged to twice normal size. Abscess about 2 cm. in left inguinal gland; abscess about 1 cm. in pectoral gland. Testicles atrophied. Pericarditis.	Organism recovered from heart blood, lung, liver, spleen, testicle, and inguinal gland.
Subcutaneous; animal 4LD	0.1 cubic centimeter of 1:1,000 dilution (1,760,000 organisms per cubic centimeter).	Died in seven days.	Pronounced necrotic lesions in lungs. Few foci in liver and spleen. Abscess about 0.5 cm. near testicle. Some orchitis present.	Organism recovered from heart blood, lung, liver, and spleen.
Subcutaneous; animal 5LD	0.5 cubic centimeter of 1:1,000 dilution.	Died in twenty-five days.	Red hepatization in lungs. Abscesses about 0.5 and 1 cm. in liver. Necrotic foci in spleen. Spleen enlarged to twice normal size. Abscess in left inguinal gland 3 cm. in diameter. Abscess 2 cm. in diameter in renal glands.	Organism recovered from heart blood, lung, liver, spleen, inguinal and renal glands.
Subcutaneous; animal 6LD	0.1 cubic centimeter of 1:100 dilution (17,600,000 organisms per cubic centimeter).	Died in nine days.	Numerous foci in liver. Pronounced lesions in lungs. Spleen enlarged to twice normal size. Old abscess about 1 cm. in diameter at point of injection. Abscess in left inguinal gland about 2 cm. in diameter.	Organism recovered from lung, liver, spleen, kidney, abscess, and intestine.

TABLE 5.—*Pathogenicity experiments on guinea pigs*—Continued.

Route of infection.	Dosage.	Result.	Post-mortem findings.	Bacteriological findings.
Nasal smear; animal 1N.	Nostrils smeared with swab dipped in 1:10 dilution of saline suspension of agar culture; Gates reading 2.1: 23–IV–25. Re-inoculation with suspension; Gates reading 3.0: 30–IV–25.	Died twenty-one days after re-inoculation.	Pronounced necrotic lesions in lungs. Few foci in liver; one large focus about 6 mm. in diameter. Pronounced lesions in spleen.	Organism recovered from lung, liver, and spleen.
Nasal smear; animal 11N	Nostrils smeared with swab dipped in broth suspension of agar culture: 25–VIII–26.	Died in eight days.	Congested pneumonic areas in lungs. Very few necrotic foci in spleen. Few foci in liver and one small necrotic area.	Organism recovered from liver.
Nasal smear; animal 12N	Nostrils smeared with swab dipped in broth suspension of agar culture: 25–VIII–26.	Died in ten days.	Congested areas throughout lungs and few necrotic foci. Numerous foci in spleen and some enlargement of spleen. Numerous foci and few necrotic areas in liver.	Organism recovered from heart blood and spleen.
Oral; animal 4F.	0.01 cubic centimeter of 1:10 dilution of broth culture: 8–I–26: 0.05 cubic centimeter: 16–I–26: 0.1 cubic centimeter: 23–I–26: 0.5 cubic centimeter: 6–II–26.	Killed moribund eight days after the last feeding.	Spleen enlarged to twice normal size. Pronounced necrotic lesions in spleen. Very few foci in lungs.	Organism recovered from spleen.
Oral; animal 5F.	0.02 cubic centimeter of 1:10 dilution of broth culture: 8–I–26: 0.05 cubic centimeter: 16–I–26: 0.1 cubic centimeter: 23–I–26: 0.5 cubic centimeter: 6–II–26: 0.5 cubic centimeter: 19–II–26: 0.5 cubic centimeter: 27–II–26.	Died five days after last feeding.	Necrotic lesions in lungs. One necrotic focus about 1 by 1 cm. in diameter and numerous pin-point foci in liver. Small abscess at sternum. Foci in spleen and some enlargement of spleen. Orchitis present.	Organism recovered from spleen and liver.

TABLE 5.—*Pathogenicity experiments on guinea pigs*—Continued.

Route of infection.	Dosage.	Result.	Post-mortem findings.	Bacteriological findings.
Oral; animal 8F.	1 cubic centimeter of serum from 2F (a fed guinea pig—to be discussed under Immunology): 23–IV–26: 0.02 cubic centimeter: 29–IV–26: 0.5 cubic centimeter: 7–V–26: 0.1 cubic centimeter: 14–V–26: 0.2 cubic centimeter: 3–VI–26: 0.5 cubic centimeter: 10–VI–26: 0.5 cubic centimeter: 1–VII–26: 0.5 cubic centimeter: 9–VII–26: 0.5 cubic centimeter: 16–VII–26: 0.75 cubic centimeter: 23–VII–26.	Died thirteen days after last feeding.	Congestion of lungs. Several necrotic foci in spleen. Numerous foci in liver.	Organism recovered from spleen, lung, and liver.
Oral; animal 10F.	0.01 cubic centimeter of 1:10 dilution of broth culture—Strain 2F: 23–IV–26: 0.02 cubic centimeter: 1–V–26: 0.05 cubic centimeter: 7–V–26: 0.1 cubic centimeter: 14–V–26: 0.5 cubic centimeter: 3–VI–26: 1 cubic centimeter: 10–VI–26.	Died four days after last feeding.	Necrotic areas in spleen and some enlargement. Foci in liver and lungs.	Do.

The bacteriological findings in the guinea pig are practically the same as those in the rabbit.

The subcutaneous and feeding methods were tested on white mice with almost as fatal results as were produced in the two species already discussed. The effects of these methods are shown in Table 6.

TABLE 6.—*Pathogenicity experiments on mice.*

Route of infection.	Dosage.	Result.	Post-mortem findings.	Bacteriological findings.
Subcutaneous; animal 1.	0.01 cubic centimeter of 1:2 dilution of saline suspension of agar culture: 18–XII–24.	Died in three days.	Numerous necrotic foci in liver. Necrotic foci in spleen.	Organism recovered from heart blood, lung, liver, and spleen.
Subcutaneous; animal 2.	0.1 cubic centimeter of 1:2 dilution of saline suspension of agar culture: 18–XII–24.	-----do-----	Necrotic foci in liver and spleen.	Organism recovered from heart blood, lungs, and spleen.
Oral; animal 3.	0.01 cubic centimeter of 1:2 dilution of saline suspension of agar culture: 18–XII–24.	Died in four days.	Very few necrotic-appearing foci in lungs. Spleen enlarged and studded with foci.	Organism recovered from heart blood, lung, liver, spleen, and kidney.
Oral; animal 4.	0.1 cubic centimeter of 1:2 dilution of saline suspension of agar culture: 18–XII–24.	Died in three days.	Necrotic foci in liver and spleen.	Organism recovered from heart blood, lung, liver, spleen, kidney, and testicle.
Subcutaneous; animal 5.	0.1 cubic centimeter of a 1:100 dilution of saline suspension of agar culture; Gates reading 1.2: 28–II–25.	Died in nine days.	Very few necrotic-appearing spots in lungs. Few foci in liver. Pronounced foci in spleen.	Organism recovered from heart blood, lung, liver, and kidney.
Subcutaneous; animal 6.	0.05 cubic centimeter of a 1:10 dilution of same suspension.	Died in four days.	Few necrotic foci in lungs. Numerous spots in liver. Pronounced necrotic lesions in spleen. Abscess about 1.5 mm. on posterior aorta.	Organism recovered from heart blood, lung, liver, spleen, kidney, and urinary bladder.
Subcutaneous; animal 7.	0.1 cubic centimeter of a 1:10 dilution of same suspension.	Eaten by other on third day.	----------	----------
Oral; animal 8.	0.1 cubic centimeter of 1:100 dilution of same suspension.	Died in eleven days.	One necrotic spot in lung. Pronounced foci in spleen and enlargement of spleen. One necrotic lesion in liver.	Organism recovered from spleen.
Oral; animal 9.	0.05 cubic centimeter of a 1:10 dilution of same suspension.	Died in five days.	Very few necrotic foci in lungs. Numerous spots in liver and numerous pronounced foci in spleen.	Organism recovered from heart blood, lung, liver, spleen, and kidney.

TABLE 6.—*Pathogenicity experiments on mice*—Continued.

Route of infection.	Dosage.	Result.	Post-mortem findings.	Bacteriological findings.
Oral; animal 10.	0.1 cubic centimeter of a 1:10 dilution of same suspension.	Died in thirteen days.	Lungs and liver normal. Numerous pronounced foci in spleen. Two abscesses about 3 mm. in diameter along spinal column.	Organism recovered from heart blood and lung.
Subcutaneous; animal 11.	0.01 cubic centimeter of 1:10 dilution of broth culture containing 52,000,000 organisms per cubic centimeter: 3-VI-26.	Died in twenty-two days.	Necrotic areas in lungs and spleen. Very few foci in liver. Necrosis at point of injection.	Organism recovered from lung, liver, and spleen.
Subcutaneous; animal 12.	0.02 cubic centimeter of 1:10 dilution of broth culture containing 52,000,000 organisms per cubic centimeter: 3-VI-26.	Died in twenty-one days.	Few foci in liver and spleen. Lungs normal.	Organism recovered from lung, liver, and spleen.
Subcutaneous; animal 13.	0.05 cubic centimeter of 1:10 dilution of broth culture containing 52,000,000 organisms per cubic centimeter: 3-VI-26.	Died in four days	Few foci in liver and spleen. Spleen enlarged to twice normal size.	Organism recovered from heart blood, liver, and spleen.
Oral; animal 14.	0.005 cubic centimeter of 1:10 dilution of broth culture containing 52,000,000 organisms per cubic centimeter: 3-VI-26.	Survived		
Oral; animal 15.	0.01 cubic centimeter of 1:10 dilution of broth culture containing 52,000,000 organisms per cubic centimeter: 3-VI-26.	Died two months later.	No lesions	Organism not recovered.
Oral; animal 16.	0.02 cubic centimeter of 1:10 dilution of broth culture containing 52,000,000 organisms per cubic centimeter: 3-VI-26.	Survived		

TABLE 6.—*Pathogenicity experiments on mice*—Continued.

Route of infection.	Dosage.	Result.	Post-mortem findings.	Bacteriological findings.
Oral; animal 17.	0.05 cubic centimeter of 1:10 dilution of broth culture containing 52,000,000 organisms per cubic centimeter: 3–VI–26.	Killed; moribund on fifth day.	Few necrotic foci in liver. Very few spots in lungs. Spleen normal.	Organism recovered from heart blood, liver, and spleen.

Loss of appetite and emaciation are the symptoms in mice. The post-mortem findings are similar to those in rabbits and guinea pigs, except that the spleen lesions seem to be more pronounced than those of the other organs, and the lungs do not show the characteristic foci as frequently as do those of the other two species. The bacteriological findings are similar also; the organism was isolated from the heart blood, lung, liver, spleen, kidney, and urinary bladder.

In two series of rats consisting of four and six animals, respectively, three of those which were fed culture died, but only one of these showed typical lesions and from only this one was the organism recovered. The effect upon these animals is shown in Table 7.

TABLE 7.—*Pathogenicity experiments on rats.*

Route of infection.	Dosage.	Result.	Post-mortem findings.	Bacteriological findings.
Subcutaneous; animal 1.	0.02 cubic centimeter of a 1:10 dilution of twenty-four hour agar culture suspended in saline; Gates 1.2: 4–II–25.	Sick on following day but recovered.	----------	----------
Subcutaneous; animal 2.	0.2 cubic centimeter of a 1:10 dilution of twenty-four hour agar culture suspended in saline; Gates 1.2: 4–II–25.	----do----	----------	----------
Oral; animal 3.	0.1 cubic centimeter of a 1:10 dilution of twenty-four hour agar culture suspended in saline; Gates 1.2: 4–II–25.	----do----	----------	----------

TABLE 7.—*Pathogenicity experiments on rats*—Continued.

Route of infection.	Dosage.	Result.	Post-mortem findings.	Bacteriological findings.
Oral; animal 4.	0.8 cubic centimeter of a 1:10 dilution of twenty-four hour agar culture suspended in saline; Gates 1.2: 4–II–25.	Sick on following day. Died four days after feeding.	Congestion and necrotic foci in the lungs. Numerous foci in the liver. Spleen slightly nodular.	Organism recovered from heart blood, lung, liver, and spleen, but not from kidney.
Subcutaneous; animal 1A.	0.02 cubic centimeter of a 1:10 dilution of broth culture—(7,000,000 organisms per cubic centimeter): 8–IX–26.	----------	----------	----------
Subcutaneous; animal 2A.	0.05 cubic centimeter of a 1:10 dilution of broth culture—(7,000,000 organisms per cubic centimeter): 8–IX–26.	----------	----------	----------
Subcutaneous; animal 3A.	0.1 cubic centimeter of a 1:10 dilution of broth culture—(7,000,000 organisms per cubic centimeter): 8–IX–26.	----------	----------	----------
Oral; animal 4A.	0.02 cubic centimeter of a 1:10 dilution of broth culture—(7,000,000 organisms per cubic centimeter): 8–IX–26.	Died five days after feeding.	Lesions not typical.	Organism not recovered.
Oral; animal 5A.	0.05 cubic centimeter of a 1:10 dilution of broth culture—(7,000,000 organisms per cubic centimeter): 8–IX–26.	----------	----------	----------
Oral; animal 6A.	0.1 cubic centimeter of a 1:10 dilution of broth culture—(7,000,000 organisms per cubic centimeter): 8–IX–26.	Died five days after feeding.	Lesions not typical.	Organism not recovered.

Increasing doses were given at irregular intervals over a period of months. In the last attempt (Table 7) 1A was given subcutaneously 1.5 cubic centimeters of a twenty-four-hour

agar culture suspended in saline; it contained approximately 104,700,000 organisms per cubic centimeter. Subcutaneous doses of 2 cubic centimeters each, were given to 2A, and 3A and 5A received 0.5 cubic centimeter per os. All the animals survived.

The effect upon the one successfully infected rat corresponds to the effect described on the other species. The organism, as the table indicates, was isolated from the heart blood, lung, liver, and spleen, but not from the kidney.

The results upon the seven pigeons tested are given in Table 8.

The two surviving birds were twice re-inoculated in the pectoral muscle with 2 cubic centimeters of a broth culture containing approximately the same number of organisms as in the previous dose. Only a slight loss of weight occurred. January 31, 1927, pigeon 5 was given intravenously a broth suspension of a twenty-four-hour agar culture; the number of organisms present was approximately 677,600,000,000 per cubic centimeter. Pigeon 6, and an uninoculated bird, 7, were given 0.5 cubic centimeter of the same suspension intravenously. Pigeon 6 lost weight, became emaciated, and developed a swelling about 1 centimeter in diameter over the right eye; a slight purulent discharge formed in the left eye. About two weeks after injection, it developed a local swelling about 0.5 by 0.8 cubic centimeter under the scales of the right leg; later this became an open ulcer (Plate 5, fig. 1). This bird was killed March 4. Post-mortem examination showed a strongly bile-pigmented liver and a mottled spleen. The lungs were normal. The eye lesion was filled with serous fluid. Neither pus nor serous fluid was observed in the leg lesion. Pigeons 5 and 7 showed no indication of sickness.

Pigeon 1 showed typical lesions. In spite of the fact that pigeon 2 showed complications, the lesions found were not unlike those produced by this organism. Pigeon 6 did not show characteristic lesions. It will be noted that the organism has been recovered from the heart blood, lung, liver, spleen, and kidney. In the case of pigeon 6, it was isolated from the eye lesion only.

The methods of inoculation tested on a chicken are given in Table 9.

TABLE 8.—*Pathogenicity experiments on pigeons.*

Route of infection.	Dosage.	Result.	Post-mortem findings.	Bacteriological findings.
Subcutaneous; animal 1.	1 cubic centimeter of twenty-four hour agar culture suspended in saline: 20–XII–24.	No indication of sickness.	----------	----------
Pricked in pectoral muscle.	Needle dipped in suspension of agar culture: 1–XII–24.	Diarrhœa. Died in three days.	Few necrotic foci on liver. Small nodules and necrotic foci in spleen.	Organism recovered from liver.
Subcutaneous.	0.5 cubic centimeter of same as above: 20–XII–24.	Diarrhœa and difficulty in standing.	----------	----------
Pricked in pectoral muscle.	0.5 cubic centimeter of same as above: 1–XII–24.	Recovered.	----------	----------
Oral; animal 2.	3 cubic centimeters of agar culture suspended in saline: 22–XII–24.	No indication of sickness.	----------	----------
	Repeated feedings to test for bacteriophage.	7–I–25. Cankers observed in the mouth. Bill was broken and was trimmed too closely and bled. Dead 12-I-25.	Area of congestion and nodules in left lung. Few purulent nodules at point of pricking.	Organism recovered from lung, liver, and spleen.
Intravenous; animal 3.	1 cubic centimeter of twenty-four hour broth culture—(2,130,000,000 organisms per cubic centimeter): 3–XII–26.	Died in twenty-four hours.	No lesions.	Organism recovered from heart blood, liver, spleen, and kidney.
Intravenous; animal 4.	------do------	-----do-----	-----do-----	Organism recovered from heart blood, lung, liver, and spleen.
Pectoral muscle injection; animal 5.	------do------	Slight loss in weight.	----------	----------
Pectoral muscle injection; animal 6.	------do------	-----do-----	----------	----------

TABLE 9.—*Pathogenicity experiments on a chicken.*

Method.	Dose.	Result.
Subcutaneous	0.4 cubic centimeter of a saline suspension of twenty-four-hour agar culture; 18–XI–24.	No indication of sickness.
Oral	1 cubic centimeter of a saline suspension of agar culture; 20–XI–24.	Do.
Do	3.5 cubic centimeters of a saline suspension of agar culture; 24–XI–24.	Do.
Intraperitoneal	2 cubic centimeters of a saline suspension of agar culture; 2–XII–24.	Do.

Toxin production.—Smith and Ten Broeck(4) report the production of toxin in filtrates of fowl typhoid bacillus grown for two days in a peptonized veal broth containing 0.1 per cent dextrose and to a less extent in beef broth. An attempt to demonstrate a toxin in two-day and fourteen-day cultures grown in meat-extract broth containing 0.1 per cent dextrose produced no effect but a slight loss in weight. By using hormone broth with a P of 8.0 to which a small amount of sterile blood was added, a forty-eight-hour culture filtrate was produced, which when introduced into the ear vein caused the death of a rabbit within twenty-four hours. The dose was 1 cubic centimeter per kilogram of weight. Post-mortem examination showed marked distension of the subcutaneous blood vessels, and congestion and apparent hæmorrhage of the subcutaneous lymph glands. The spleen was engorged and enlarged; the parenchyma on the cut surface was bulging and jamlike. Extensive mottled areas occurred in the liver; there appeared to be parenchymatous degeneration. A slightly blood-stained exudate was present in the peritoneum. The lungs showed hypostatic congestion, and the thymus gland contained hæmorrhagic spots.

Twenty-three days after the first rabbit was inoculated, a second rabbit was given the same filtrate by the same route. A rise in temperature occurred within a few hours, but the temperature dropped to normal the following morning. A rabbit receiving intravenously a fourteen-day culture filtrate in the same medium showed a rise of temperature within a few hours. In this case, also, the temperature dropped the following day. Loss of weight was observed in both cases. This seems to indicate that some toxin is produced.

PRELIMINARY NOTES ON IMMUNOLOGY

The extreme virulence of this organism makes immunization a difficult problem and no real success has been obtained.

Agglutinin production: Six normal rabbits were given intravenous injections of a 1:5 dilution of forty-eight-hour culture suspended in physiological saline containing 5 per cent formalin. The inoculations were made on three successive days, using a dose of 0.25, 0.5, and 1 cubic centimeter. A week later, the animals were bled and their serums tested for the presence of agglutinins.

The results obtained are given in Table 10.

TABLE 10.—*Agglutination tests on rabbits.*[a]

[Antigen: 1:5 dilution of stock saline suspension of forty-eight-hour culture containing 5 per cent formalin]

Donor of serum.	Serum dilutions.						
	1:10	1:20	1:40	1:80	1:160	1:320	1:640
Rabbit 1	++	++	++	±	∓	—	—
	++	++	++	+	±	∓	—
Rabbit 2	++	++	++	±	∓	∓	—
	++	++	++	+	±	∓	∓
Rabbit 3	++	++	++	∓	∓	—	—
	++	++	++	±	±	∓	—
Rabbit 4	++	++	++	++	++	∓	—
	++	++	++	++	++	±	—
Rabbit 5	++	++	++	++	∓	—	—
	++	++	++	++	±	∓	—
Rabbit 6	++	++	++	++	++	∓	—
	++	++	++	++	++	±	—

[a] First reading, after overnight incubation.
Second reading, after twenty-four hours, standing at room temperature.
— indicates no evidence of reaction.
± indicates slight sediment but supernatant fluid turbid.
∓ indicates more sediment than ±, but still a faint cloudiness in supernatant fluid.
++ indicates complete agglutination after overnight incubation.
+ indicates incomplete agglutination after overnight incubation, but a clearing up of the fluid upon standing for twenty-four hours.

To increase the titer of these serums, the animals were re-inoculated with the same doses at weekly instead of daily intervals. Rabbit 3 died after the first re-injection, and, because of a loss in weight, rabbit 5 was given only one re-injection. The tests following these injections gave the results shown in Table 11.

It will be observed from this table that the highest definite titer obtained is 1:320, indicating that agglutinin production is not very strong.

TABLE 11.—*Results of repeated inoculations to increase the titer of agglutination serums.*

[Antigen: 1:50 dilution of stock saline suspension of forty-eight-hour culture containing 5 per cent formalin.]

Donor of serum.	Serum dilutions.													
	1:10	1:20	1:40	1:80	1:100	1:120	1:160	1:200	1:240	1:320				
Rabbit 1	++	++	++	++	++	++	++	±	∓	—				
	++	++	++	++	++	++	++	+	∓	∓				
	1:10	1:20	1:40	1:80	1:160	1:200	1:240	1:320	1:400	1:480	1:640	1:800	1:960	1:1280
Rabbit 2	++	++	++	++	++	++	++	++	±	±	—	—		
	++	++	++	++	++	++	++	++	±	+	∓	—		
Rabbit 4	++	++	++	++	++	++	++	++	∓	∓	∓	—	—	—
	++	++	++	++	++	++	++	++	±	∓	∓	∓	∓	—
Rabbit 5	++	++	++	++	++	++	++	++	∓	∓	∓	—		
	++	++	++	++	++	++	++	++	∓	∓	∓	—		
	1:10	1:20	1:40	1:80	1:160	1:320	1:640	1:1280						
Rabbit 6	++	++	++	++	++	++	++	±						
	++	++	++	++	++	++	++	+						
							1:640	1:800	1:960	1:1280				
							±	±	±	—				
							±	±	±	—				

The rabbits used for agglutinin production were tested with living cultures to find out whether there was any immunity present. This experiment was carried out almost two months after the last injection of killed culture, but the serums tested previous to this time showed practically the same titer. The results obtained are shown in Table 12.

TABLE 12.—*Tests for immunity in rabbits in which agglutins were present.*

Route of infection.	Dosage.	Result.	Post-mortem findings.	Bacteriological findings.
Intraveneous; animal R–1.	0.5 cubic centimeter of 1:100 dilution of broth culture containing 85,000,000 organisms per cubic centimeter.	Died in forty-eight hours.	Numerous necrotic foci in liver and spleen; one necrotic spot in the lung.	Organism recovered from heart blood, liver, spleen, and kidney.
Subcutaneous; animal R–2.	0.5 cubic centimeter of broth suspension.	Died in seventy-two hours.	Necrotic foci in liver and spleen.	Organism recovered from heart blood, lung, liver, spleen, and uterus.
Subcutaneous; animal R–4.	0.2 cubic centimeter of broth suspension.	---do----	-----do----------	Organism recovered from liver and spleen.
Oral; animal R–5.	0.5 cubic centimeter of broth suspension.	Died on thirty-fourth day.	Necrotic foci in lungs, liver, and spleen; very few in kidney.	Organism recovered from spleen.
Oral; animal R–6.	1 cubic centimeter of broth suspension.	Survived---	----------	----------
Subcutaneous; animal 7 (control).	0.2 cubic centimeter of broth suspension.	Died on fourth day.	Typical lesions------	Organism recovered from heart blood, lung, liver, spleen, kidney, testicle, and urinary bladder.

Table 12 indicates that only one rabbit of the series showed any immunity; since there was no control on the fed animal, this survival does not indicate much.

An attempt to produce an unattenuated vaccine was abandoned when it was discovered that 0.1 cubic centimeter of a 1:100,000 dilution of broth culture was lethal for a rabbit.

Repeated subcutaneous inoculations at one- or few-day intervals of a heat-killed suspension of culture were tried on a rabbit. Fourteen injections were given in one case. Eight days after the last injection, a lethal dose was given subcutaneously and the rabbit succumbed in six days. This experi-

ment was repeated with a suspension killed in 1 per cent trikresol. A week following the ninth injection, a living suspension was inoculated subcutaneously into the rabbit and the animal died in three days.

An attenuated vaccine was prepared by grinding to a paste the livers and spleens of infected rabbits and diluting this with physiological saline containing glycerin to the amount of one-third its volume. The P_H of this glycerin-saline solution was 7.8. Glycerin equivalent to one-third the total volume was then added and enough phenol to make a 0.5 per cent suspension in the mixture. The vaccine was heated in a water bath at 44 to 46° C. for three hours. Two guinea pigs were injected subcutaneously with 1 cubic centimeter doses at three successive weekly intervals. A week following the last inoculation, one of these animals was given a 0.2 cubic centimeter dose; the other, a 0.1 cubic centimeter dose of a 1 : 100 dilution of twenty-four-hour agar culture suspended in saline. The two control guinea pigs received the same respective doses. All the animals died; apparently the vaccine had no effect.

So-called artificial aggressins were prepared according to the method of Citron(5) by shaking aqueous and serous suspensions of agar cultures for forty-eight hours, centrifuging, and preserving the supernatant fluid. One rabbit received subcutaneously 2.5 cubic centimeters of serous aggressin; one received 3 cubic centimeters of aqueous aggressin. On the thirty-second day, each animal, together with a control rabbit, was given a lethal dose of culture. The control and the rabbit receiving the serous aggressin died on the fourth day following injection. The rabbit receiving the aqueous aggressin succumbed on the fifth day, showing that no immunity was produced.

Experiments to produce immunity by feeding living cultures are shown in Table 13.

TABLE 13.—*Experiment to produce immunity in a guinea pig by feeding increasing doses of live bacteria.*

Animal 2-F:

Received 0.2 cubic centimeter of a 1 : 100 dilution of agar culture suspended in saline; 24-XI-25.

Received 0.2 cubic centimeter of a 1 : 10 dilution of broth culture; 5-XII-25.

Received 0.1 cubic centimeter of a 1 : 10 dilution of broth culture; 12-XII-25.

Received 0.1 cubic centimeter of a 1 : 10 dilution of broth culture; 19-XII-25.

TABLE 13.—*Experiment to produce immunity in a guinea pig by feeding increasing doses of live bacteria*—Continued.

Animal 2–F—Continued.
Received 2 cubic centimeters of a 1 : 10 dilution of broth culture; 24–XII–25.
Received 2 cubic centimeters of a 1 : 10 dilution of broth culture; 7–I–26.
Received 2.5 cubic centimeters of a 1 : 10 dilution of broth culture; 16–I–26.
Received 0.5 cubic centimeter of undiluted broth culture; 23–I–26.
Received 0.5 cubic centimeter of broth culture subcutaneously; 6–II–26.
Control:
Received 0.5 cubic centimeter of broth culture subcutaneously; 6–II–26.

The control pig died in six days (Plate 5, fig. 2); and the fed animal acquired an abscess at the point of injection. It was reinoculated with 1 cubic centimeter of broth culture twelve days after the first injection, became sick, and died nine days later. Autopsy showed an old abscess at the point of inoculation; an abscess about 1 centimeter in diameter was present in the inguinal gland, and very few necrotic foci were observed in the spleen. The organism was recovered from the inguinal abscess.

The second attempt is given in Table 14.

TABLE 14.—*Experiment to produce immunity in a guinea pig by feeding increasing doses of live bacteria.*

Animal 13:
Received 0.2 cubic centimeter of broth culture (6,000,000 organisms per cubic centimeter); 26–VIII–26.
Received 0.5 cubic centimeter of broth culture; 2–IX–26.
Received 0.1 cubic centimeter of broth culture; 7–IX–26.
Received 0.2 cubic centimeter of broth culture; 16–IX–26.
Received 0.5 cubic centimeter of broth culture; 23–IX–26.
Received 1.0 cubic centimeter of broth culture; 30–IX–26.
Received 1.5 cubic centimeters of broth culture; 7–X–26.
Received 2 cubic centimeters of broth culture; 14–X–26.
Received 0.5 cubic centimeter of broth culture (540,000,000 organisms per cubic centimeter) subcutaneously; 22–X–26.
Control:
Received 0.5 cubic centimeter of broth culture subcutaneously; 22–X–26.

The control animal died six days after the injection. Guinea pig 13 developed a large abscess around the point of inoculation (Plate 6); it lived twenty-three days. Post-mortem findings

showed typical lesions in both pigs, and the organism was recovered. The temperature curve for these animals is shown in text fig. 6.

It appears from these experiments that a very low-grade immunity may be produced by feeding increasing doses of living culture.

DISCUSSION

The following bacterial incitants have been incriminated or associated with pneumonic lesions in swine: *Bacillus suisepticus* (Zinsser and Russell),(6) or *Pasteurella suisepticus* in

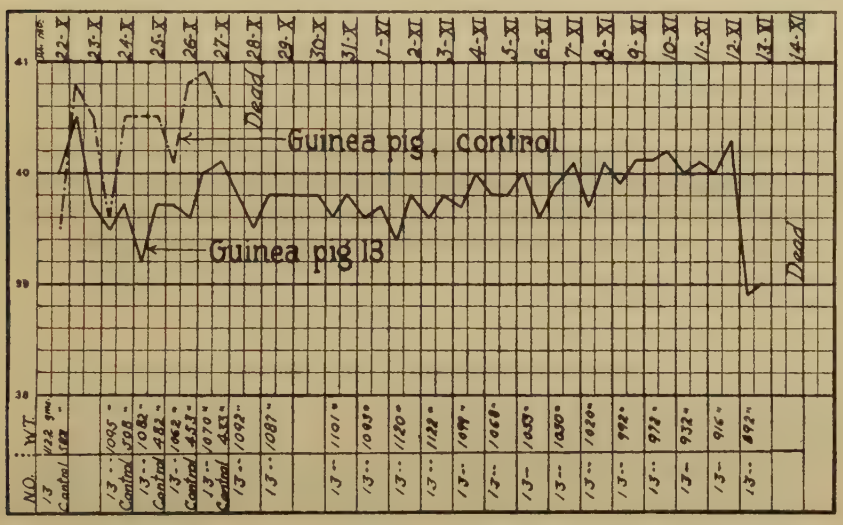

FIG. 6. Temperature curve of two guinea pigs which received subcutaneously 0.5 cubic centimeter of broth culture containing 540,000,000 organisms per cubic centimeter. Guinea pig 13 had previously been given oral doses of live bacteria in an attempt to immunize it.

Bergey's(7) classification, is the causative agent of swine plague, a disease very rarely encountered in the United States. Briefly, the organism is a short, oval, Gram-negative, bipolar, nonmotile rod, belonging to the hæmorrhagic septicæmia group. This same organism is looked upon as a secondary invader in hog cholera.

In a similar manner, *Bacillus suipestifer* (Hutyra and Marek),(8) or *Salmonella suipestifer* (Bergey),(7) is associated with hog cholera and has also been isolated from pneumonia in hogs. This organism is a motile rod, possessing peritrichous flagella; it is negative to Gram stain and belongs to the paratyphoid group.

In swine erysipelas, caused by *Bacillus rhusiopathiae suis* (Hutyra and Marek),(8) or *Erysipelothrix rhusiopathiae* (Bergey);(7) inflammatory changes are also present. *Bacillus rhusiopathiae suis* (Buchanan)(9) is a very slender rod with a tendency to form filaments; it is nonmotile, takes the Gram stain, and is grouped by Bergey in the genus *Erysipelothrix.*

Swine are also susceptible to lung changes produced by the nonmotile, acid-fast tubercle bacillus, *Mycobacterium tuberculosis* (Bergey).(7)

In enzoötic pneumonia, or so-called chronic swine plague, Hutyra and Marek(8) claim that a number of agents are involved. Aside from *Bacillus suisepticus,* which, though frequently present, is considered not to be the primary etiologic agent, *Bacillus pyogenes suis* has been found often. This, as the name indicates, is a pus-producing, weakly Gram-positive, slender, nonmotile rod (Hutyra and Marek).(8) The fluorescent Gram-negative, motile rod, *Bacillus pyocyaneus* (*Pseudomonas aeruginosa* Bergey),(7) is associated with his disease, and the well-known colon bacillus, streptococci, and staphylococci have also been found.

SUMMARY AND CONCLUSION

Here is described a taxonomic study of a microörganism isolated by Dr. William H. Boynton from pneumonic lesions of hogs in the Philippine Islands.

This organism was found to be a Gram-negative motile rod, measuring 1.2 to 3.6 microns in length, and 0.5 to 1.5 microns in thickness, and possessing 1 to 5 monotrichous flagella. This indicates that it belongs to the genus *Pseudomonas.* It liquefies gelatin; coagulates and peptonizes milk; produces neither indol nor hydrogen sulphide; reduces nitrates to nitrites; produces acid but no gas in dextrose and galactose; produces a slight transcient acidity in mannose, arabinose, maltose, mannite, dulcite, and glycerin, but no fermentation in levulose, saccharose, lactose, xylose, or dextrin. It is highly pathogenic for rabbits, guinea pigs, and mice, slightly so for pigeons, and very slightly pathogenic for rats.

Unsuccessful attempts to immunize rabbits and guinea pigs are described.

Since a search of the available literature (Bergey(7), Chester(10), Migula(11), Levine(12)) failed to reveal an organism which corresponds with this one, apparently this organism has not been described, and the name *Pseudomonas suis* sp. nov. is proposed for it.

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ILLUSTRATIONS

PLATE 1

A chronic case in a guinea pig, injected with 0.5 cubic centimeter of broth culture on January 11, 1927. Photograph was taken January 24. Death occurred on January 29. Metastatic abscess, *a;* metastatic abscesses becoming open ulcers, *b* and *c;* abscess at point of injection becoming an open ulcer, *d.*

PLATE 2

Liver lesions in a guinea pig. Pinpoint necrotic focus, *a;* large focus, *b.*

PLATE 3

Lung lesions in a guinea pig. Necrotic foci, *a* and *b;* pronounced necrotic lesion, *c.*

PLATE 4

Spleen lesions in a guinea pig. Small focus, *a;* larger lesion, *b.*

PLATE 5

FIG. 1. Pigeon 6. Eye swelling, *a;* leg ulcer, *b.*

2. Contrast in appearance between Guinea pig 2F and its sick control. Guinea pig 2F was fed increasing weekly doses of live organisms before receiving a subcutaneous dose.

PLATE 6

Guinea pig 13 which was fed increasing weekly doses of live culture before receiving a subcutaneous dose. Ulcer at point of injection is shown.

TEXT FIGURES

FIG. 1. The swine-pneumonia organism, showing flagella. (After Pitfield.)

2. Growth curve of the organism.
3. Temperature curve of two hogs which were fed livers, hearts, spleens, and lungs of two guinea pigs infected with the swine-pneumonia organism, 30–VIII–23.
4. Temperature curve of a rabbit which received subcutaneously 0.2 cubic centimeter of a 1 : 10 dilution of 16-hour broth culture containing approximately 85,000,000 organisms, 4–XII–26.
5. Temperature curve of two guinea pigs inoculated by smearing their nostrils with a swab dipped in a broth suspension of a 24-hour agar culture, 25–VIII–26.
6. Temperature curve of two guinea pigs which received subcutaneously 0.5 cubic centimeter of broth culture containing 540,000,000 organisms per cubic centimeter. Guinea pig 13 had previously been given oral doses of live bacteria in an attempt to immunize it.

PLATE 1.

PLATE 2.

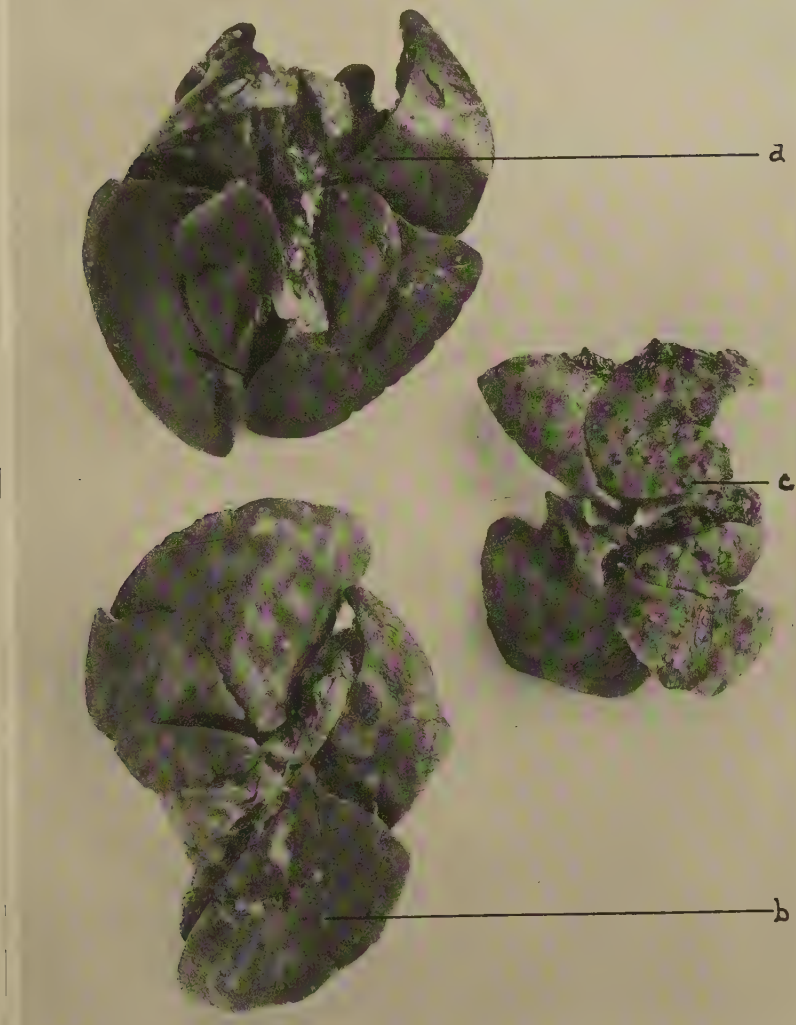


PLATE 3.

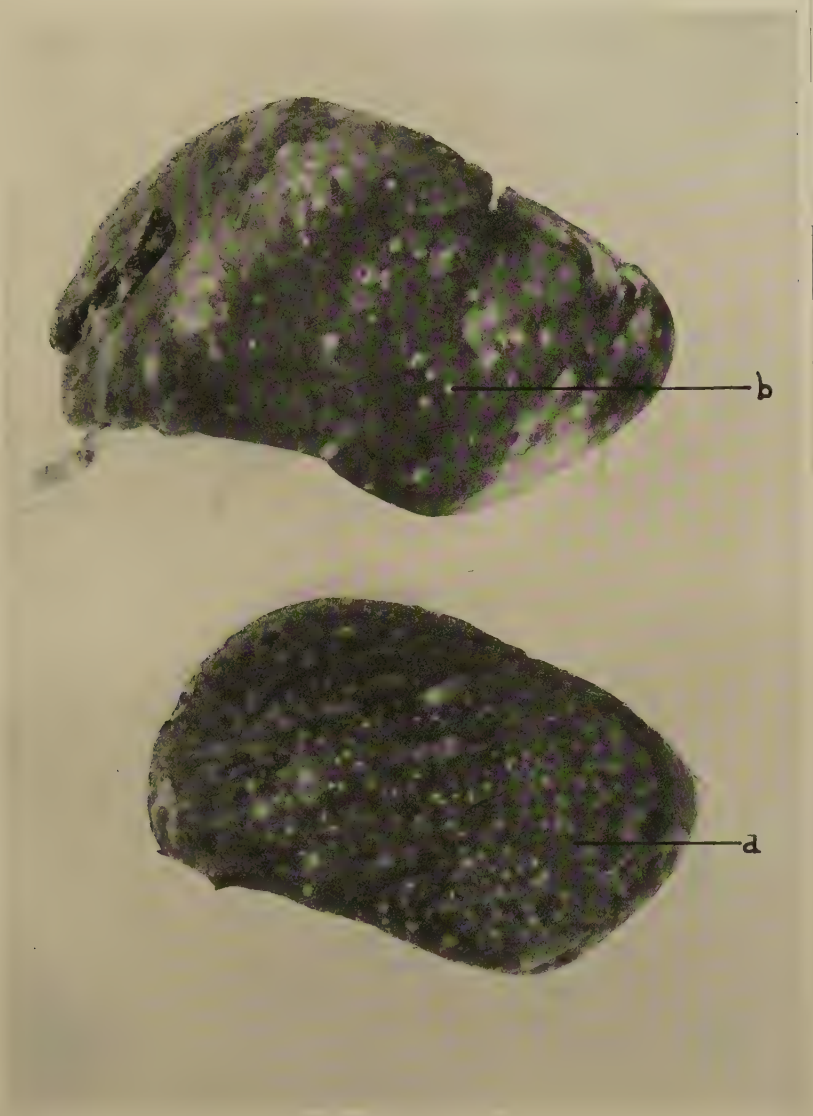
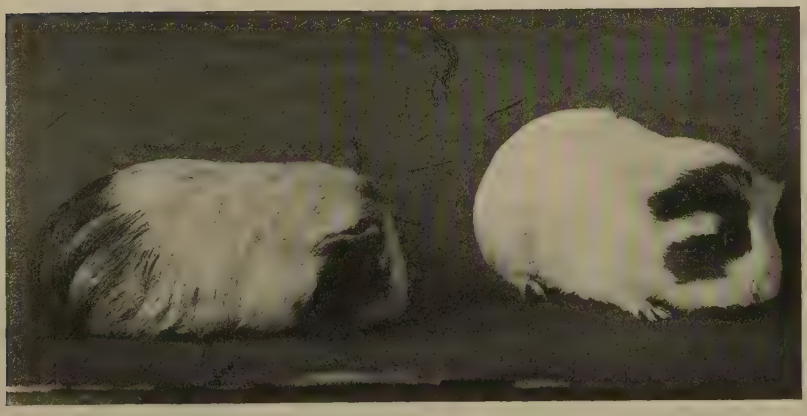


PLATE 4.

1



2

PLATE 5.

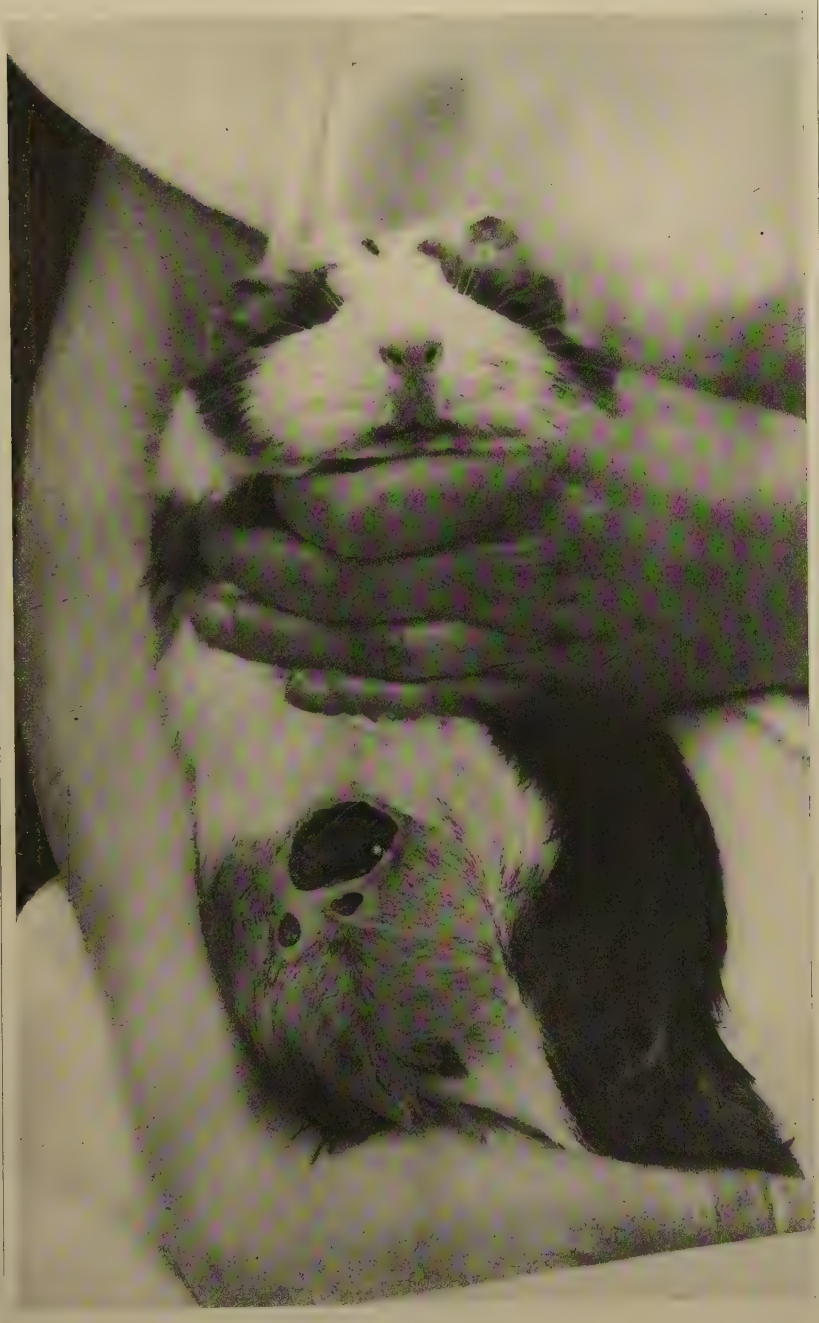


PLATE 6.

TRANSMISSION OF DENGUE FEVER BY AËDES ALBOPICTUS SKUSE[1]

By James Stevens Simmons
Major, Medical Corps, United States Army
Joe H. St. John
Major, Medical Corps, United States Army
and
Francois H. K. Reynolds[2]
Captain, Veterinary Corps, United States Army

ONE PLATE AND ONE TEXT FIGURE

Though a number of different mosquitoes have been incriminated as possible dengue vectors the only species generally accepted without question during the past few years is *Aëdes ægypti.* This mosquito was used experimentally for the transmission of dengue in 1906 by Bancroft,(1) in 1916, 1918, and 1919 by Cleland, Bradley, and MacDonald,(2) and in 1923 by Chandler and Rice;(3) and its remarkable effectiveness in this respect was conclusively demonstrated in 1926 by Siler, Hall, and Hitchens.(4) However, those who have worked with *A. ægypti* have not claimed that it was the only carrier of dengue. On the other hand certain observers have suggested the possibility that other species, including *Aëdes* (*Stegomyia*) *albopictus* Skuse (*Stegomyia scutellaris* Walker), might also transmit the disease. For example, in 1923 Cleland(5) in an article on dengue observed that, "Some epidemiological evidence suggests that *Stegomyia scutellaris* Walker, may also be a vector;" yet it appears that this possibility was not considered seriously, since in the same paper he stated that, "The disease is conveyed by *Aëdes* (*Stegomyia*) *fasciata* and perhaps by this mosquito alone." *Aëdes albopictus* was also suspected by Koizumi, Yamaguchi, and Tonomura(6) during the 1915 dengue epidemic in Formosa, and was used by these investigators in transmission

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[2] Assisted by Staff Sergeant Jesse Rhodes, M. D., Sergeant Carl Heilmann, M. D., Private 1st. cl. Hinton Miller, M. D., and Private 1st cl. Bonifacio Reyes, M. D. (P. S.).

experiments. Unfortunately, as was pointed out by the authors, their results were inconclusive because the human volunteers used had not been previously protected against accidental mosquito bites, and therefore the part played by *A. albopictus* in the dissemination of dengue remained unproved.

In view of the suggestive but inconclusive evidence just mentioned, and observations which showed *A. albopictus* to be not uncommon in Manila during the recent dengue season, it was considered of importance to settle the question concerning its ability to transmit dengue to man.

AËDES (STEGOMYIA) ALBOPICTUS SKUSE [3]

Aëdes (*Stegomyia*) *albopictus* Skuse is the name now generally applied to *Stegomyia scutellaris* Walker, which has also been known by the following synonyms: *Culex scutellaris* Walker (1859); *Culex variegatus* Doleschall (1858) (non Schrank 1781) (non Blanchard 1852); *Culex albopictus* Skuse (1895); *Stegomyia scutellaris* Theobald (1901); *Stegomyia scutellaris samarensis* Ludlow (1911); and *Stegomyia albopicta* Skuse.

This mosquito was described in 1901 by Theobald(7) as follows:

3. STEGOMYIA SCUTELLARIS. Walker (1859).

C. scutellaris. Walker.
C. albopictus. Skuse.
C. variegatus. Doleschall.

* * * * * * *

Thorax black, with a median silvery stripe; abdomen with silvery white basal bands; legs black, tarsi basally white banded, last joint of the hind legs pure white.

♀. Head with a patch of dark scales on each side, separated by a broad band of silvery-white scales, which form a bright spot in front, and with a silvery-white border to the eyes, and another patch on each side of the head; eyes partly black and golden; antennae dark brown, faintly paler banded at the joints, with a tuft of silvery scales on the basal joints, forming two distinct spots; palpi black with a silvery white apical joint; proboscis black.

[3] Banks(9) in 1906 advanced the supposition that *S. scutellaris* might be closely related to *S. fasciata* and that some intergradation might possibly occur. However, this has not been accepted by Theobald,(7) Barraud,(8) Edwards,(10) or members of this board who have observed that *A. albopictus* is an easily distinguished species. Moreover, carefully controlled experiments conducted in these laboratories show that *A. albopictus* and *A. ægypti* failed to interbreed, thus eliminating any question concerning the specific identity of these two mosquitoes.

Thorax black, covered with black scales, and fine, rather dull brown, hairs; a distinct, clear, silvery line in the middle from the front to about two-thirds of the way across the mesonotum; scutellum edged with silvery scales; metanotum dark brown; pleurae black with numerous silvery-white spots. Abdomen covered with black scales, the bases of the segments with a band of silvery scales which spread out laterally, most noticeable on the fourth and fifth segments, forming distinct lateral patches; ventral surface also with white scales.

Legs black, the femora with silvery knee spots and pale beneath for part of their length; tibiae black; metatarsi with broad white basal bands; in the fore and mid feet the first tarsal joint is basally white, the others black; in the hind legs all the tarsi with broad, white, basal bands, except the last, which is pure white. Front claws untoothed. Wings with the veins covered with long brown scales and a double row of short darker ones; first sub-marginal cell short, slightly longer and narrower than the second posterior cell; posterior cross-vein some distance behind the mid cross-vein. Halteres with pale testaceous stem and dusky knob.

Length.—4.5 to 5 mm.

♂. Thorax, abdomen, and legs like the ♀. Antennae banded black and white, the basal joint black with a large patch of silvery-white scales on the inside; plumes deep brown; palpi long and thin, not plumed, black, with two broad white bands towards the base and a white spot underneath at the base of the last two joints; proboscis black; fore and mid ungues unequal, the larger with one large tooth, the smaller simple; hind ones small and simple, equal.

Length.—4 to 4.5 mm.

Habitat.—Singapore (Raffiesian Museum) (4. 9. 1899); Hong Kong (Ford) (27. 9. 1899); Selangor (A. L. Butler) (28. 10. 1899); Upper Burma (Watson); North Borneo; Mauritius (Sir Charles Bruce) (22. 11. 1899); Tamsui, Formosa (Mackay) (2. 8. 1899); Fiji (Black) (30. 12. 1899); Japan (Wood); Celebes (Walker); Ceylon (Bartholomew) (12. 12. 1899); Madras and Naini Tal, India (Giles and Cornwall); Siam (Skeate); Amboina (Doleschall); Sombalpur, C. P., India (D. O'C. Murphy) (99); Foo-Chow, China (Rennie) (9. 8. 1900) (84).

Time of capture.—Singapore, July (July 27, 1899); Ceylon, November; Upper Burma (March).

Observations.—This is a very common mosquito, with a wide distribution in Asia. It is a common species in the Straits Settlements, being the second commonest mosquito in Selangor (A. L. Butler). Ford records its larvae as being abundant "in standing water near houses 500 feet above the sea." It is a great nuisance at Calcutta (Skuse). Skeate also evidently took it in abundance in Siam, for numbers are in the collection sent me by Dr. Sharpe.

* * * * * * *

Synonymy.—Skuse's *C. albopictus*, described in the Indian Museum Notes, iii. 5, p. 20, is certainly this species, every character agreeing with Walker's type of *C. scutellaris*, which occurs commonly in India. Doleschall's *Culex variegatus* is evidently the same as *C. scutellaris;* Doleschall described it from Amboina, where it is one of the most troublesome mosquitoes throughout the year, common in houses.

In 1922 Barraud(8) published the following description of *A. albopictus* in which he called special attention to the distinctive form of the male hypopygium:

Stegomyia albopicta (Skuse).

Culex albopictus, Skuse, Ind. Mus. Notes iii, No. 5, p. 20. (1895).

Stegomyia scutellaris, Theobald (nec Walker), Mon. Cul. vol. i, p. 298. (1901).

Stegomyia samarensis, Ludlow, Psyche, vol. xviii, p. 127. (1911).

* * * * * * *

The only other known Indian species of *Stegomyia* with which this may be confused is *unilineata*, but the absence of white spots on the mesonotum, and on the mid femora, of *albopicta*, form fairly easily seen distinctions.

The male hypopygium is somewhat like that of *S. argentea* (*fasciata*), but from the drawings it will be seen that there are differences in the shape of the clasper, ventro-lateral plate of the anal segment, and in the ninth tergite.

There are specimens in the Central Malaria Bureau collection from the following places:—

North-West Frontier:—Kohat (Sinton).

Punjab:—Simla and Delhi (Christophers). Kasauli (Christophers and Sinton). Amritsar (Barraud).

Bombay:—Islands in the harbour, and Trombay (Barraud).

Bombay Deccan:—Belgaum and Nagargali (Barraud).

Malabar Coast and Nilgiri Hills (Khazan Chaud).

Madras Carnatic:—Madras (Patton). Salem (Christophers).

Bihar:—Cuttack (S. Sundar Rao).

Burma:—Rangoon (Christophers).

The following additional localities are taken from Theobald (Mon. Cul.):—Calcutta (Annandale); Upper Burma (Watson); Madras and Naini Tal (Giles and Cornwall); Sombalpur, Cen. Prov. (D. O'C. Murphy); Kanara district (E. H. Aitken); Sylhet, Assam (Major Hall); Lushai Hills, Assam (E. C. Macleod); Manipur (C. A. Gourlay); Katihar and Purneah district; N. Bengal (C. A. Paiva); Ceylon (Green).

PRESENT INVESTIGATION

This investigation was begun by collecting larvæ of different species of *Aëdes* in order to learn which ones were numerically important in Manila. Since the results obtained indicated that next to *A. ægypti* the commonest mosquito of this genus was *A. albopictus*, the latter was bred in the laboratory and used experimentally for the transmission of dengue.

PREVALENCE OF A. ALBOPICTUS IN THE PHILIPPINE ISLANDS

A review of the literature shows that *A. albopictus* Skuse has been reported as present in the Philippine Islands by a number of observers. Ludlow(11) in 1903 described it under the name

Stegomyia scutellaris samarensis; and again in 1908 she(12) remarked that in places where *Stegomyia calopis* was infrequent its place was largely taken by *Stegomyia scutellaris.* Banks(9) in 1906 reported that he had failed to collect *S. scutellaris* Walker, but that *S. scutellaris samarensis* Ludlow, "seemed a widespread mosquito in the Philippines." He named the following locations in which *S. scutellaris* had been found: Pangasinan, Camp Gregg, Bayambang (Col. W. P. Chamberlain, M. C., U. S. Army), Samar, Leyte, Mindoro, Iloilo, Occidental Negros, Bago (Banks), Manila, Fort William McKinley (Col. Charles F. Craig, M. C., U. S. Army), and various collectors. Since the terms *S. scutellaris* Walker and *S. scutellaris samarensis* Ludlow are now accepted as synonyms for *A. albopictus* Skuse, it is apparent that this common oriental mosquito is widely distributed in the Philippine Archipelago.

The attention of the board was first directed to *A. albopictus* in February, 1929, when specimens were observed in a small lot of mosquitoes which had been caught on Corregidor Island, Philippine Islands. More recent collections of *Aëdes* larvæ have been made between April 12 and September 1, 1929, in the city of Manila. The results as indicated in Table 1 show that larvæ of this mosquito were often found in the same breeding places with *A. ægypti,* and that the relative proportion of *A. albopictus* increased during June and July at which time dengue fever was very prevalent.

TABLE 1.—*Relative prevalence of A. albopictus Skuse and A. ægypti in Manila, as indicated by collection of larvæ.*

Month.	Number of samples.	Samples containing *A. albopictus.*	Predominant species.	Monthly admissions of dengue-fever patients at Sternberg General Hospital Manila.
		Per cent.		
April	1	100	*A. ægypti*	18
May	3	100	do	16
June	4	75	*A. albopictus*	38
July	16	100	Neither	33
August	104	43	*A. ægypti*	11

DENGUE TRANSMISSION BY A. ALBOPICTUS

The dengue virus.—The "K" strain of dengue virus used in testing the ability of *A. albopictus* to transmit the infection was

obtained December 15, 1928, by feeding normal female *A. ægypti* on an American soldier (Keller) who had developed dengue fever a short time previously. Since that date the "K"

CHART 1.—*Transfer of "K" strain of dengue virus from December 15, 1928, to July 16, 1929, by Aëdes ægypti.*

Keller, dengue fever
Second day, December 15, 1928.

↓

Lot 1, *Aëdes ægypti*
17 fed December 31, 1928.

↓

American volunteer 1 (H), dengue fever
First day, January 10, 1929.

↓

Lot 3, *Aëdes ægypti*
56 fed February 19, 1929.

↓

American volunteer 3 (A), dengue fever
First day, February 25, 1929.

↓

Lot 9, *Aëdes ægypti*
49 fed April 11, 1929.

↓

American volunteer 6 (R), dengue fever
First day, April 16, 1929

↓

Lot 12, *Aëdes ægypti*
15 fed May 8, 1929.

↓

American volunteer 9 (D), dengue fever
First day, May 14, 1929.

↓

Lot 19, *Aëdes ægypti*
12 fed June 20, 1929.

↓

American volunteer 14 (A), dengue fever
First day, June 26, 1929.

↓

Lot 25, *Aëdes ægypti*
14 fed July 11, 1929.

↓

American volunteer 22 (M), dengue fever
First day, July 16, 1929.

↓

See Chart 1*a*.

strain of virus has been employed extensively in various other experiments; and as indicated in Chart 1, the particular branch of this strain used since July 16, 1929, for the *A. albopictus* transmission experiments had previously been passed alternately through six different lots of *A. ægypti* and six human volunteers.

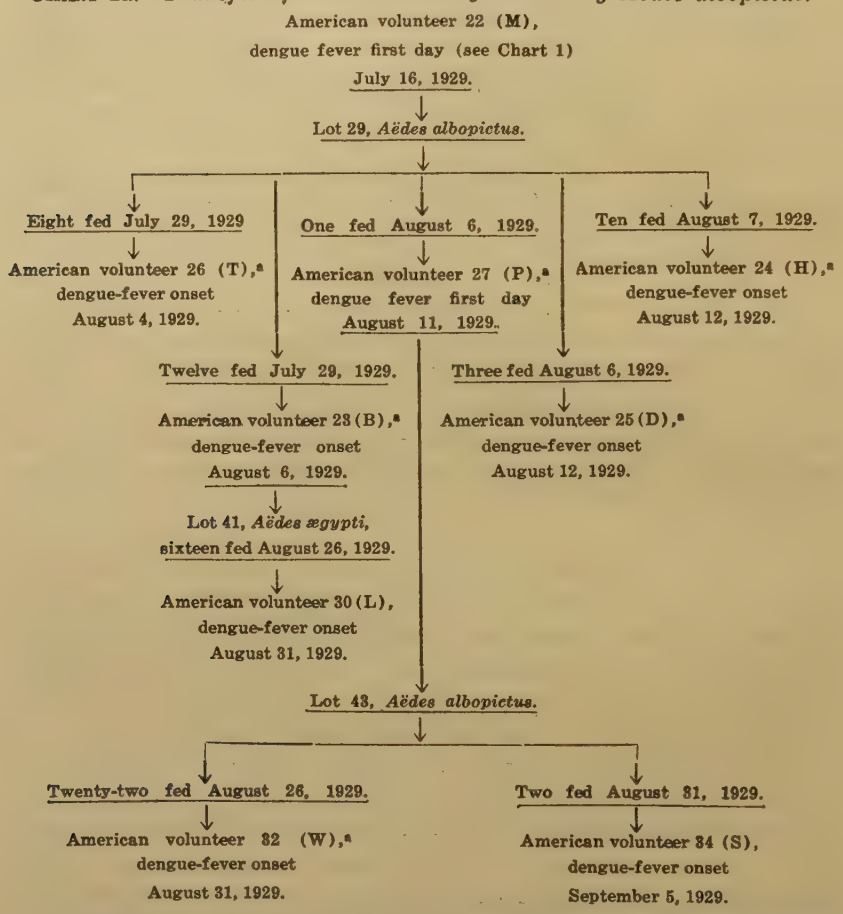
Aëdes albopictus.—The mosquitoes selected had emerged from larvæ collected about Manila, and from larvæ descended from a carefully selected stock bred in the laboratory. The specific identity of each mosquito used either for breeding purposes or in the transmission experiments was determined by a number of individual examinations made by members of the board at the following times: Immediately after emergence from the pupal stage; before being placed in the breeding cages; before and after taking an infective blood feeding from a dengue case; and before and after feeding on volunteers. Specimens from one of the experimental lots were also submitted to the Chief of the Bureau of Entomology, Washington, D. C., whose report confirmed the identification of the mosquito as *A. albopictus* Skuse. Two lots of mosquitoes designated as 29 and 43 were used in the transmission experiments.

Aëdes albopictus lot 29.—One hundred female mosquitoes recently emerged from larvæ, some of which had been collected outside and others of which had been bred in the laboratory, were placed in a cage of the type devised by Siler, Hall, and Hitchens(4) for feeding mosquitoes on human beings. July 16, 1929, these mosquitoes were given an opportunity to feed on a volunteer patient (American volunteer 22–M) who had developed typical symptoms of dengue fever twelve hours earlier. Eighty mosquitoes immediately settled on the exposed skin and within a very short time they were all engorged with blood.

These potentially infected *A. albopictus* were designated as lot 29, and kept for a minimum period of thirteen days before tests were made to determine their ability to transmit dengue to man. Small groups consisting of 1, 3, 8, 10, and 12 mosquitoes, respectively, were fed on each of five human volunteers (see Chart 1*a*). Since these men had previously been used for other experimental dengue inoculations which had resulted negatively, the positive results obtained served not only to prove that the volunteers were susceptible to dengue, but also indicated the infectivity of the mosquitoes. Each volunteer had been isolated in a screened cubicle inside the screened ward used by the board

for dengue investigations at the Sternberg General Hospital, Manila (Plate 1), for eight to ten days previous to the experiments and for additional periods of two or more weeks before being used in these final tests. In every instance an interval of eight or more days was allowed between the last negative experiment and the feeding of *A. albopictus* to rule out the possibility of infection resulting from the former procedure within the usual incubation period.

CHART 1a.—*Transfer of "K" strain dengue virus by Aëdes albopictus.*


American volunteer 22 (M),
dengue fever first day (see Chart 1)
July 16, 1929.
Lot 29, Aëdes albopictus.
Eight fed July 29, 1929
American volunteer 26 (T),a dengue-fever onset August 4, 1929.
One fed August 6, 1929.
American volunteer 27 (P),a dengue fever first day August 11, 1929.
Ten fed August 7, 1929.
American volunteer 24 (H),a dengue-fever onset August 12, 1929.
Twelve fed July 29, 1929.
American volunteer 23 (B),a dengue-fever onset August 6, 1929.
Lot 41, Aëdes ægypti, sixteen fed August 26, 1929.
American volunteer 30 (L), dengue-fever onset August 31, 1929.
Three fed August 6, 1929.
American volunteer 25 (D),a dengue-fever onset August 12, 1929.
Lot 43, Aëdes albopictus.
Twenty-two fed August 26, 1929.
American volunteer 32 (W),a dengue-fever onset August 31, 1929.
Two fed August 31, 1929.
American volunteer 34 (S), dengue-fever onset September 5, 1929.

a Volunteers 23, 24, 25, 26, 27, and 32 previously had been used experimentally in negative attemps to produce dengue fever, and *A. albopictus* was employed to test their immunity to dengue.

As shown in the protocols the five volunteers (Nos. 23, 24, 25, 26, and 27) developed typical dengue fever within five to seven days after they were bitten by infected *A. albopictus* of lot 29. These observations were further confirmed by transferring the

infection through new lots of mosquitoes to still other human volunteers. In the case of volunteer 23, this was done by feeding normal *A. ægypti* during the first day of dengue fever and passing the infection on from these mosquitoes to a second volunteer (American volunteer 30–L). From another case (American volunteer 27–P), which developed after the bite of one mosquito of lot 29, the virus was transferred to a new lot of *A. albopictus* (lot 43) which later transmitted the infection to two other human volunteers (American volunteers 32–W and 34–S).

Aëdes albopictus lot 43.—A group of fifty-four female mosquitoes, descendants of the original stock which had bred in the laboratory for several generations, were infected August 11, 1929, by feeding on the blood of American volunteer 27–P, who twelve hours previously had developed typical symptoms of dengue. Since this case of dengue had resulted from the bite of a single mosquito of lot 29, the new potentially infected group of lot 43 mosquitoes represented a second transfer of the virus by *A. albopictus.*

After periods of fifteen and twenty days, respectively, small groups of the lot 43 mosquitoes were tested on two volunteers. In the first instances twenty-two mosquitoes were employed to prove the susceptibility of a man (American volunteer 32–W) who had been used previously for another experiment. The other volunteer (American volunteer 34–S) was admitted to the ward for the sole purpose of determining the ability of *A. albopictus* to transmit dengue, and was isolated in a screened cubicle for ten days, after which he was bitten by two mosquitoes of this lot. Five days later he developed dengue fever (see protocol 7).

The seven successful transmission experiments reported here indicate that *A. albopictus* is highly susceptible to infection with dengue virus, and prove conclusively that it is an effective dengue vector.

PROTOCOLS

EXPERIMENT 1

American volunteer No. 23 (Beaudrot), N. J., Pvt. Co. K, 31st Inf., age 22 years, white, service 2 months. Born and lived in Charleston, S. C. Arrived in Philippine Islands June 15, 1920. No history of dengue. Admitted to experimental ward June 22, 1929 (No. 101086). Physical examination and routine laboratory tests showed him to be normal. *Preliminary observation period* nine days, from June 22 to July 1, 1929. No evidence of dengue. *Negative experiments.* Inoculated subcutaneously with Mandler filtrates of saline suspension of infected *A. ægypti* on July 1, 11, and 20, all of which failed to produce dengue. *Test for susceptibility to dengue.* Twelve female *A. albopictus* (lot 29), which had been infected

13 days previously from an experimental dengue case (Am. vol. 22–M) took blood feedings on July 29, 1929. *Dengue fever.* Onset 7 days later, the morning of Aug. 5, 1929. Symptoms typical including rash at onset, with fever, leucopenia, weakness, pains in eyes, knees, and elbows, and an itching terminal rash on Aug. 10, after which temperature remained normal. During the first day, Aug. 6, 1929, *A. ægypti* were fed on this case and used later to produce dengue fever in another volunteer (Am. vol. 30–L). *Conclusions.* The feeding of 12 infected *A. albopictus* (lot 29) was followed after 7 days by typical dengue fever.

EXPERIMENT 2

American volunteer No. 24 (Heist), Pvt. Co. I, 31st Inf., age 25 years, service 3 months. Born in Pottstown, Pa. Arrived in Philippine Islands June 15, 1929. No history of dengue. Admitted to experimental ward June 27, 1929 (No. 101129). Physical examination negative; history of probable luetic infection one year ago; Wassermann reaction positive. Otherwise normal. *Preliminary observation period,* 13 days, from June 27, to July 11, 1929. No evidence of dengue. *Negative experiments.* Inoculated subcutaneously (1) July 11, 1929 with Mandler filtrate of saline suspension infected *A. ægypti;* (2) on July 21, with suspension of filtered four day old dried dengue blood; and (3) on July 29, bitten by *Culex* mosquitoes which had previously taken blood from a case of dengue. Since all of these procedures failed to produce dengue fever, the volunteer's susceptibility was tested by feeding infected *A. albopictus,* on August 7. *Test for susceptibility* to dengue. Ten female *A. albopictus* (lot 29) which had been infected 22 days previously from an experimental dengue case (Am. vol. 22–M) took blood feedings on Aug. 7. *Dengue fever.* Onset 5 days later, Monday, August 12. Attack began abruptly with chills, followed by fever, headache, generalized pains, and flushed face. The typical symptoms including pain of back and eyes, and leucopenia persisted until Aug. 15. Discharged Aug. 20, 1929. *Conclusions.* The feeding of 10 infected *A. albopictus* (lot 29) was followed after 5 days by typical dengue fever.

EXPERIMENT 3

American volunteer No. 26 (Tolson), Pvt. Co. A, 31st Inf., age 23 years, white. Arrived in Philippine Islands June 15, 1929. No history of dengue. Admitted to experimental ward June 29, 1929 (No. 101158). Physical examination and routine laboratory tests showed him to be normal. *Preliminary observation period,* 12 days, from June 29, to July 11, 1929. Normal. No evidence of dengue. *Negative experiments.* On July 11, 1929, 2 infected *A. ægypti* were killed and rubbed on the unbroken skin; and on July 21, 0.5 cc. of a saline suspension of unfiltered four days old dried dengue blood was injected subcutaneously. Both of these procedures failed to produce dengue. *Test for susceptibility to dengue.* Eight female *A. albopictus* (lot 29) which had been infected 13 days previously from an experimental dengue case (Am. vol. 22–M) took blood feedings on July 29, 1929. *Dengue fever.* Onset 6 days later, in the afternoon of Aug. 4, 1929, with fever, leucopenia, and a generalized rash. Had typical symptoms of rather marked severity which lasted 4 days. *Conclusions.* The feeding of 8 infected *A. albopictus* (lot 29) was followed in 6 days by typical dengue fever.

EXPERIMENT 4

American volunteer No. 25 (Dandrea), Pvt. Co. C, 31st Inf., white, age 18 years, service 2 months. Born in Vermont. Arrived in Philippine Islands June 15, 1929. No history of dengue. Admitted to experimental ward June 29, 1929 (No. 101157). Physical examination and routine laboratory tests showed him to be normal. *Preliminary observation period,* 12 days, from June 29, to July 11, 1929. No evidence of dengue. *Negative experiments.* On July 11, injected subcutaneously with Mandler filtrate of infected *A. ægypti;* on July 21, injected subcutaneously with Mandler filtrate of infected *A. ægypti;* and on July 29, 4 *Culex* mosquitoes previously fed on a dengue case took blood. *Test for susceptibility to dengue.* Three female *A. albopictus* (lot 29) which had been infected 21 days previously on an experimental dengue case (Am. vol. 22–M) took blood feeding on Aug. 6, 1929. *Dengue fever.* Onset 7 days later, on the morning of Aug. 12, 1929, with typical symptoms of a mild case without any initial rash. The infection lasted 5 days, ending with a terminal rash which appeared Aug. 17, 1929. *Conclusions.* The feeding of 3 infected *A. albopictus* (lot 29) was followed in 7 days by typical dengue fever.

EXPERIMENT 5

American volunteer No. 27 (Pringle), Pvt. Co. 28, Bomb sqdn., Nichols Field, P. I., white, age 19 years, service 3 months. Born and lived in New Jersey. No history of dengue. Admitted to experimental ward July 2, 1929 (No. 1011812). Physical examination and routine laboratory tests showed him to be normal. *Preliminary observation period,* 9 days, from July 2, to July 11, 1929. No evidence of dengue. *Negative experiments.* On July 11, two infected *A. ægypti* were killed and rubbed on scratched skin of forearm; on July 21, injected subcutaneously with Mandler filtrate of lot 29 *A. albopictus;* and on July 29, injected with Mandler filtrate of infected *A. ægypti.* All of these procedures failed to produce dengue. *Tests for susceptibility.* On Aug. 6, one *A. albopictus* (lot 29) which had been infected 21 days previously on an experimental dengue case (Am. vol. 22–M) took a full feeding of blood. *Dengue fever.* Onset 5 days later, on the afternoon of Aug. 11, 1929, with fever, leucopenia, anorexia, vomiting, dizziness, headache, and pains in eyes and muscles. On the second day a typical rash appeared, and general glandular enlargement was noted later in the disease. The infection lasted for 5 days. On the first day of dengue fever (Aug. 11) normal *A. albopictus* designated as lot 43 were given a feeding of blood. Two cases of dengue (experiments 6 and 7) were later produced by these lot 43 mosquitoes. *Conclusions.* The feeding of one infected *A. albopictus* (lot 29) was followed in 5 days by typical dengue fever.

EXPERIMENT 6

American volunteer No. 32 (Whittier), Pvt. 66th Service Squadron, Nichols Field, P. I., white, age 21 years. Born in Oakland, Cal. No history of dengue. Arrived in Philippine Islands April, 1929. Admitted to experimental ward Aug. 9, 1929. Physical examination and routine laboratory tests showed him to be normal. *Preliminary observation period,* 8 days, from Aug. 9, to Aug. 17, 1929. No evidence of dengue. *Negative experiments.* Aug. 17, injected subcutaneously with 52-day-old desiccated

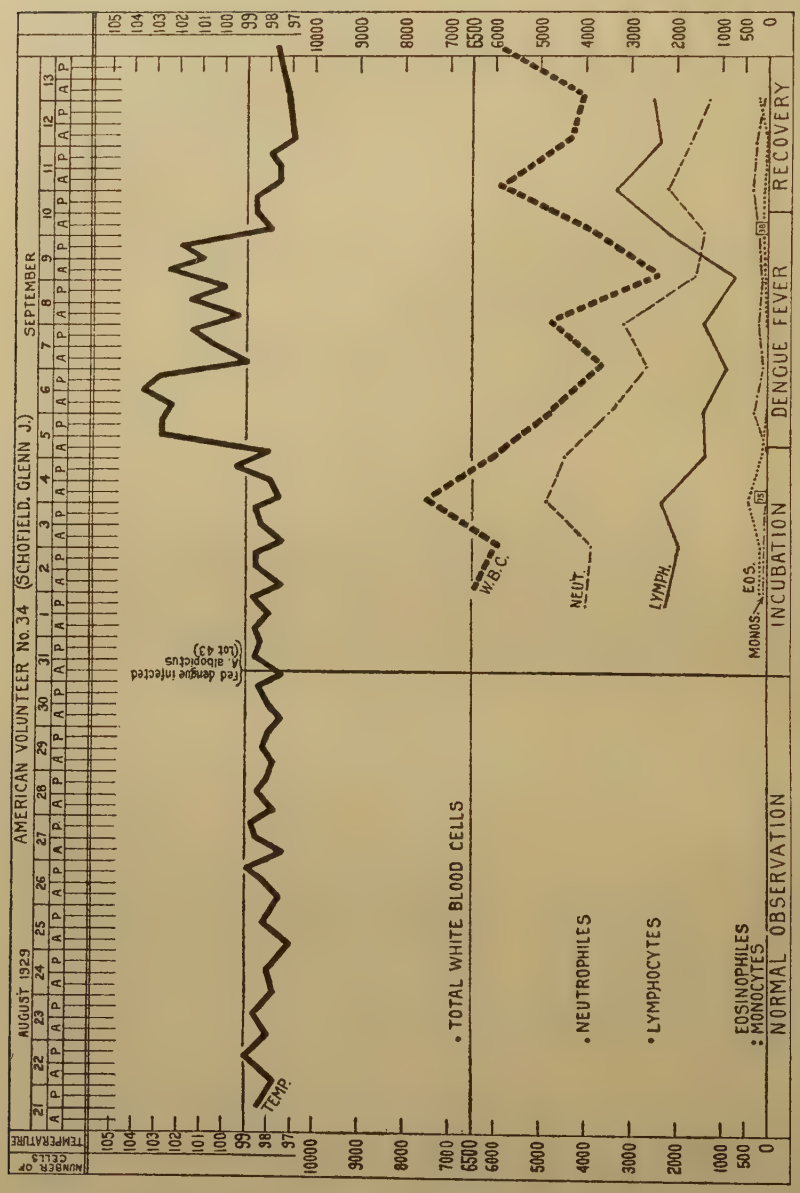


FIG. 1. Dengue fever transmitted by *Aedes albopictus*. Daily temperature and leucocyte record of volunteer Schofield, before, during, and following dengue fever produced by the two *A. albopictus* of lot 43.

dengue blood. Dengue did not develop. *Tests for susceptibility.* Twenty-two *A. albopictus* (lot 43) which had fed 15 days previously on experimental dengue case (Am. vol. 27–P) took full feeding of blood Aug. 26, 1929. *Dengue fever.* Onset 5 days later, on Aug. 31, 1929, with headache, nausea, fever, and leucopenia. The duration of the disease was 5 days, with generalized pains, nausea and vomiting, and dizziness. There was no rash. *Conclusions.* The feeding of 22 infected *A. albopictus* (lot 43) was followed after 5 days by typical dengue fever.

EXPERIMENT 7

American volunteer No. 34 (Schofield) Pvt. Detachment, M. D., Nichols Field, P. I., white, age 23 years, born in Montana, and has lived in Oregon and Idaho. No history of dengue. Arrived in Philippine Islands April, 1929. Admitted to experimental ward Aug. 21, 1929. Physical examination and routine laboratory tests showed him to be normal. *Preliminary observation period,* 10 days, from Aug. 21, to Aug. 31, 1929. *Transmission experiment with A. albopictus.* Two *A. albopictus* (lot 43) previously infected from experimental dengue case (Am. vol. 28–P) were fed on this volunteer August 31, 1929. *Dengue fever.* Onset 5 days later, on Sept. 5, with headache, pain in eyes and muscles, weakness, dizziness, generalized rash, fever and leucopenia. Duration of the disease was 5½ days. *Conclusions.* The feeding of 2 infected *A. albopictus* (lot 43) was followed in 5 days by typical dengue fever, proving that this mosquito is an effective vector.

DISCUSSION

The results of the present investigation which prove that *A. albopictus,* a common oriental mosquito, is an effective dengue carrier make it necessary to reconsider the available evidence concerning the epidemiological importance of this species. From the observations made by the board and by others who have studied the mosquito, it is apparent that *A. albopictus* somewhat resembles *A. ægypti* in its choice of breeding places and in its habit of feeding on human beings mainly during the day, although the former mosquito may require a few more days for development to the adult stage, and appears to be even more vicious and daring in biting man. The geographical distribution of *A. albopictus* given by Theobald,(7) Barraud,(8) and others indicate that this species is prevalent in many localities, including India, Formosa, the Philippine Islands, and other parts of the Orient where dengue fever is endemic. Cleland(5) and others mention the fact that *A. albopictus* has been suspected as a dengue vector on epidemiological grounds alone; while this mosquito was under grave suspicion during the 1915 dengue epidemic in Formosa.(6) Furthermore, observations made in Manila during the present year indicate that this mosquito was especially prevalent during June and July at the time

of the highest incidence of dengue fever. From the evidence presented in this report it is obvious that *A. albopictus* must be considered seriously as a factor in the dissemination of dengue fever.

SUMMARY AND CONCLUSIONS

1. Recent collections of larvæ indicate that *Aëdes albopictus*, like *A. ægypti*, is a common mosquito in Manila.

2. Female *A. albopictus* bred in the laboratory were shown experimentally to be effective transmitters of dengue from man to man. Seven human volunteers, bitten by 1, 2, 3, 8, 10, 12 and 22 *A. albopictus*, respectively, which had been infected between thirteen and twenty-two days previously, all developed dengue fever.

3. *Aëdes albopictus* must be considered as an agent in the dissemination of dengue.

4. In view of the fact that two distinct species of *Aëdes* are now known to be dengue vectors it is reasonable to suppose that still others may be concerned in the spread of this disease.

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ILLUSTRATIONS

PLATE 1. Screened cubicles for isolation of human volunteers used for dengue-transmission experiments, in screened ward, Sternberg General Hospital, United States Army, Manila, Philippine Islands. (Photographed by United States Army Signal Corps.)

TEXT FIGURE

FIG. 1. Dengue fever transmitted by *Aëdes albopictus*. Daily temperature and leucocyte record of volunteer Schofield, before, during, and following dengue fever produced by the two *A. albopictus* of lot 43.

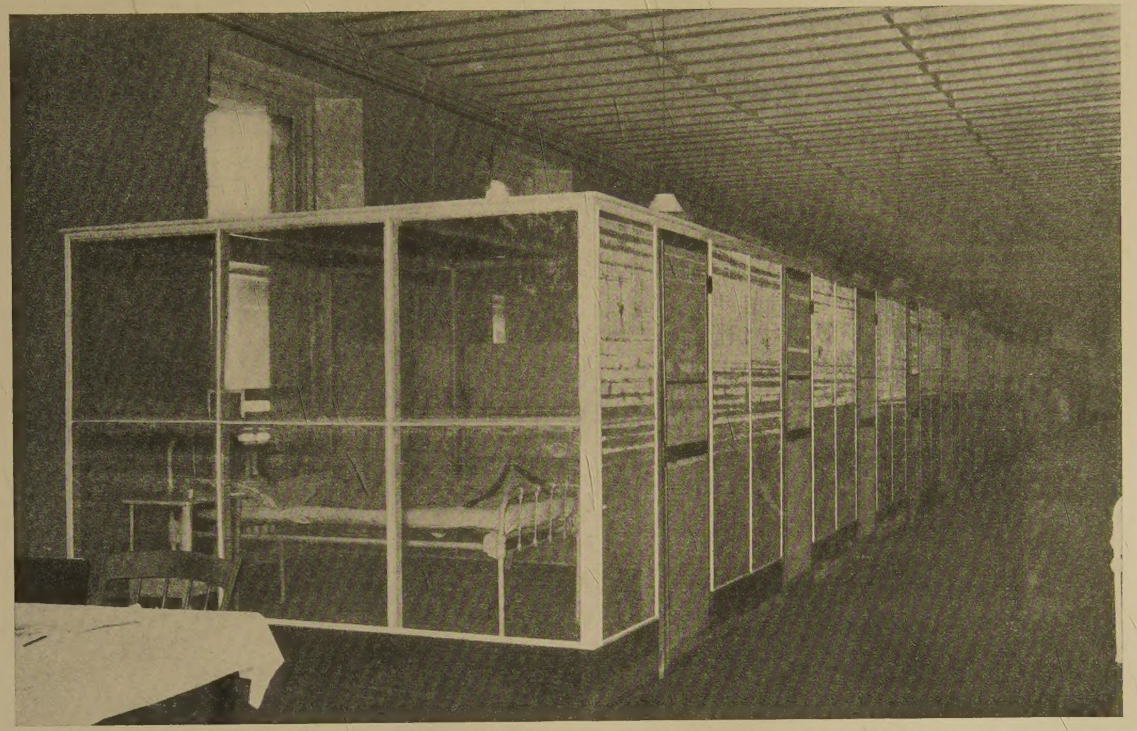

PLATE 1. SCREENED CUBICLES FOR ISOLATION OF HUMAN VOLUNTEERS.
(Photographed by United States Army Signal Corps.)